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The Safe Drinking Water Act of 1974 provides a unique opportunity for the Federal Government to constructively add to the traditional and varied roles of state and local governments in providing safe drinking water for this Nation's public. This volume focuses on the present standards for coliform bacteria, coliform bacteria detection and control, compliance with the coliform standard, and alternative means of determining compliance, and lays the groundwork for future regulations development. It is anticipated that the discussions in this proceedings, in addition to the National Academy of Sciences Review, will provide the Environmental Protection Agency with new insights into the technical problems associated with the M.C.L.'s for coliform bacteria and help to establish a firm basis for their revision.

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EVALUATION OF
THE MICROBIOLOGY STANDARDS
FOR DRINKING WATER

Edited by

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U.S. ENVIRONMENTAL PROTECTION AGENCY
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PREFACE

Although the quality of America's drinking water is often taken for granted, safe drinking water is a result of careful utilization of the latest technology and proper water treatment. In the late 1800's it was discovered that microorganisms responsible for diseases such as typhoid and cholera could pass through the intestinal tract and find their way to the water systems. By 1900, 30 people per 100,000 population died of typhoid in the United States. The death rate from typhoid fever was substantially reduced with the addition of filtration to water supply facilities and many cities had this equipment on line by 1907. Chlorination was being practiced in most U.S. cities by 1914, and during the period 1946–1970 waterborne outbreaks of typhoid fever had dropped to a total of 53.

It is apparent that the incidence of typhoid fever and cholera has been drastically reduced in the United States, but there is also information to indicate that waterborne disease warrants further investigation. While there were 53 cases of typhoid during the years of 1946–1977, there were 297 outbreaks of gastroenteritis of undetermined etiology. It is also significant to note that 71 percent of these outbreaks occurred from private water supplies but at least 83 percent of the illnesses were associated with community water systems.

The above statistics indicate a risk of infectious disease associated with drinking water, but they do not address whether the water supplied met the 1962 Public Health Service Drinking Water Standards. It is significant to note that one of the reasons for the passage of the Safe Drinking Water Act of 1974, was the lack of uniform application of the 1962 Standards to all public water systems. Another largely unknown factor is the extent to which the present microbiology standards are adequate to protect against waterborne disease. Certainly the epidemiological data supports the adequacy of the standards for protection against cholera and typhoid but the prevalence of the less severe diseases such as hepatitis, giardiasis and gastroenteritis suggests that some adjustment of the regulations may be required. This volume focuses on the present standards for coliform bacteria, coliform bacteria detection and control, compliance with the coliform standard, and alternative means of determining compliance, and lays the groundwork for future regulations development.

Sincere appreciation is acknowledged for the effort of Dr. Riley D. Housewright, Mr. Edwin E. Geldreich and Dr. S.M. Morrison for aiding in the planning of the symposium. Special acknowledgments are also given to Cindy Moneymaker and Lynette Simmons for their secretarial support and to Barbara Menking for her editorial assistance in preparing this Proceedings.

Charles W. Hendricks
August 1978

FOREWORD

The Safe Drinking Water Act of 1974 provides a unique opportunity for the federal government to constructively add to the traditional and varied roles of State and local governments in providing safe drinking water for this nation's public. Included in the Safe Drinking Water Act are requirements for the promulgation of Primary Drinking Water Regulations which will apply to all public water supplies. These regulations are to contain:

1. a list of contaminants that have adverse effects on human health.
2. maximum contaminant levels (MCL's).
3. criteria and procedures to assure a supply of drinking water which dependably complies with such maximum contaminant levels.

On December 24, 1975, the U.S. Environmental Protection Agency promulgated National Interim Primary Drinking Water Regulations. These Regulations replaced the Public Health Service Drinking Water Standards of 1962 and for the first time applied maximum contaminant levels for microorganisms and selected chemicals to all public water supplies serving 25 or more persons. It is estimated that there are at least 50,000 community and more than 200,000 non-community supplies covered by the Interim Primary Regulations.

The papers in this Proceedings focus on current microbiological issues and problems associated with the coliform MCL and the means of determining compliance. It is anticipated that these discussions, in addition to the National Academy of Sciences review, will provide the Environmental Protection Agency with new insights into the technical problems associated with the MCL's for coliform bacteria and help to establish a firm basis for their revision.

Joseph A. Cotruvo
Director
Criteria and Standards Division
Office of Drinking Water (WH-550)
U.S. Environmental Protection Agency

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WELCOME

Dr. Joseph A. Cotruvo, Director
Criteria and Standards Division
Office of Drinking Water
U.S. Environmental Protection Agency
Washington, D.C. 20460

I want to extend a sincere welcome to all of you in attendance at today's symposium entitled "Evaluation of the Microbiology Standards for Drinking Water". We are fortunate to have many of the nation's leading experts in water supply who will address both the technical and regulatory aspects of the current regulations. I would invite all of you to participate in the discussions to the maximum extent possible and everyone here should find something of interest.

The symposium is sponsored by the Criteria and Standards Division, Office of Water Supply and is intended to gather information in support of future regulations development. The Safe Drinking Water Act (PL-93-523) requires EPA to revise the Interim Primary Regulations within 90 days after the receipt of the National Academy of Science review. The presentations and discussions here will add to our understanding of the problems and issues and, together with the National Academy of Sciences review, will enable us to make the necessary revisions to the regulations.

Despite the advanced technology and management practices applied to water treatment, waterborne disease outbreaks still occur and a significant portion of the time we cannot determine what etiological agent is involved. Most of the outbreaks and cases of illness resulted from contamination of groundwater and deficiencies in treatment but many of the outbreaks (31%) were due to inadequacies or interruptions in the chlorination process. This indicates that we must constantly improve the operation of existing treatment and monitoring processes as well as consider new analytical methods and techniques.

The preamble of the Interim Primary Regulations addressed three specific areas where changes in the primary regulations may be desirable. The monthly average by which compliance is determined, is affected by the upper limit of coliform bacteria when the sample contains coliform bacteria too numerous to count. For example, some laboratories assume an upper limit of 50 while other laboratories seek to count all the colonies on the plate. There is also a problem in dealing with spurious positive samples. All samples taken to determine compliance must be counted because a negative check sample taken days after a positive sample does not mean that the original positive result was in error. The third aspect to be resolved concerns relating monthly averages of coliform bacteria to monthly percentages of positive samples. Each routine sample taken during the month may be in compliance with the limit and a system could still fail the monthly average maximum contaminant level. I would anticipate, in your discussions over the next two days, each of these questions could be discussed in detail as to provide the Criteria and Standards Division with a means of dealing with them without compromising the integrity of the present maximum contaminant levels.

To help facilitate answering these and other compelling questions, the symposium is divided into three sessions. Session 1–Evaluation of the Coliform Standard, will have Dr. Riley D. Housewright from the National Academy of Sciences, Safe Drinking Water Committee, to act as chairman. His session will focus on microbial water quality, alternative indicators, and the problems encountered with testing procedures and treatment. Session 2–Coliform Detection and Control, chaired by Mr. Edwin E. Geldreich, Municipal and Environmental Research Laboratory, Cincinnati, Ohio, will deal with the problems of the growth and detection of coliform bacteria and other indicator organisms. In addition, viruses, turbidity and water reuse will be discussed. Dr. Charles W. Hendricks from my office will chair Session 3–

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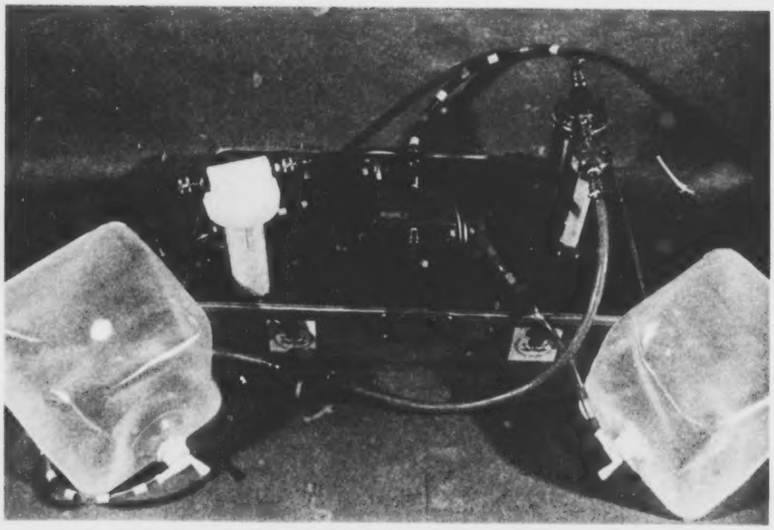
Compliance with Coliform Standards. This discussion includes the compliance aspects of the National Interim Primary Regulations and specific changes that should be made in the primary regulations. A panel discussion to be conducted by Dr. Sumner M. Morrison from Colorado State University will consider alternative means of determining compliance with bacteriological standards.

I thank all of you for attending and hope this symposium will be beneficial for all concerned as we work toward providing safe drinking water.



Fly-Away Virus Concentrator (US Environmental Protection Agency)

SESSION 1

Evaluation of the Coliform Standard

Chairman: Dr. Riley D. Housewright, Project Director, Safe Drinking Water Committee, National Academy of Sciences, Washington, D.C.

Evaluating the Microbial Quality of Potable Waters

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Bacterial Indicator Concepts

Coliform bacteria have been the bacteriological tool used to measure the occurrence and intensity of fecal contamination in drinking waters for nearly 70 years (11,13). During this period, a mass of microbiological data has been developed to permit the conclusion that the absence of total coliforms in a potable water supply is evidence of a safe supply. Although there are some differences between various coliform strains with regard to natural survival and perhaps resistance to disinfectants, these are minor biological variations that are more clearly demonstrated in the laboratory than in a water treatment system. The presence of any coliform bacteria, fecal or nonfecal, in treated water should not be tolerated, and when found, suggests either inadequate treatment or postchlorination contamination in the distribution system (5,15). For these reasons it is not surprising that the total coliform indicator system is part of this nation's drinking water standards.

The coliform group of bacteria was first adopted in 1914 by the U.S. Public Health Service pursuant to the Interstate Quarantine Regulations for common carriers (21). Since the inclusion of this bacterial quality parameter for potable waters, the coliform concept has remained essentially unchanged in successive Public Health Service standards of water quality, i.e. 1925, 1942, 1946. During this time there have been a few minor refinements/clarifications in the multiple tube procedure. The development and acceptance of the membrane filter analytical method in the 1962 Public Health Service Standards added an alternate procedure, but since then no further advances have been made in methodology. Most recently, with the promulgation of the National Interim Primary Drinking Water Regulations in 1975, the total coliform group of bacteria was again selected as the indicator of sanitary significance for evaluating the microbial quality of drinking water (20). In view of advances in methodologies for detection of enteric viruses and bacterial pathogens, the infallibility of the coliform standard has been occasionally questioned (18) although it continues to be the basic biological test of water potability.

To enumerate all waterborne pathogenic organisms, the microbiologist would have to perform a variety of complex, time-consuming, costly, and often tentative procedures for each sample analyzed. A more realistic approach is to use a bacterial indicator system which will detect and quantify fecal pollution from all warm-blooded animals. Insofar as bacterial pathogens are concerned, the total coliform group of organisms, which comprise only a fraction of the number and types of bacteria present in feces (12), is considered a reliable indicator of the adequacy of water treatment. From a practical standpoint, the standard total coliform methods (MPN, MF) offer the finished water laboratory a microbial test parameter which is easy to perform, requires little time, modest equipment expenditures, is relatively inexpensive, and provides results in 24 hours for the MF or within 96 hours when using the MPN procedure. Both the MPN and MF protocols can be easily learned and performed by most water treatment plant personnel.

While the coliform standard and its concomitant methodology are the preferred microbial surveillance tool, there have been instances where such monitoring has not signaled the presence of waterborne *Giardia* cysts. Newly reported data on non-filtered surface water systems note that the removal of coliforms during the disinfection process may not be an effective measure of cyst inactivation (9). Data

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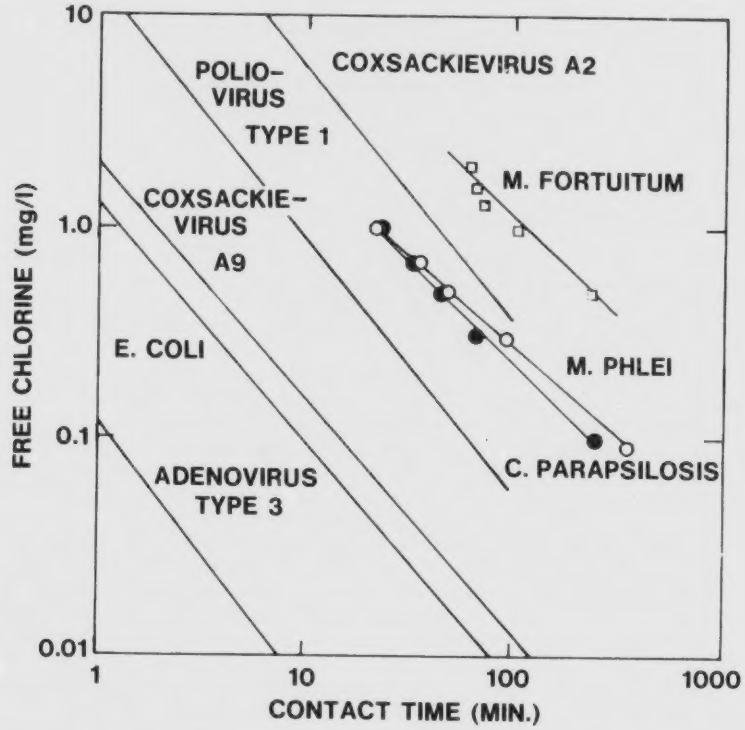

Figure 1. FREE CHLORINE RESIDUALS AND CONTACT TIMES NECESSARY FOR 99.9% INACTIVATION FOR SELECTED VIRUSES, BACTERIA, AND YEAST (2, 10).

collected to correlate virus occurrences with total coliforms in finished water support the continued use of the coliform group as the sanitary standard.

The Relative Resistance of Bacteria and Viruses

There is a considerable amount of information available on the relative resistance of bacteria and viruses to chemical disinfectants. Although a number of viruses may occur in polluted surface waters, i.e. infectious hepatitis virus, poliovirus, reovirus, adenovirus, echovirus, coxsackievirus and bacteriophages, disinfection data are limited principally to the enteric viral groups of polio and coxsackievirus (16). Because chlorine is widely used as the disinfectant of choice in most water treatment plants, determination of the relative resistance of these enteric viruses to free chlorine and chloramines is of obvious practical importance. Ideally, it is desirable that the general resistance of all enteric viruses to chlorination would be of the same order and be similar to inactivation rates of the indicator coliform bacteria. The literature on studies using a variety of microorganisms and species of chlorine/chloramines show that there are measurable differences in susceptibility to disinfectants (Figure 1). While some enteric viruses, bacteria (*Mycobacterium fortuitum, M. phlei*), and yeast (*Candida parapsilosis*) are more resistant that coliforms to inactivation by conventional disinfectants (10), current recommended water treatment practices use chlorine concentrations greater than 1 mg/l with protracted contact periods to increase the likelihood of total inactivation of viral and bacterial pathogens.

The Significance of Virus-Coliform Ratios

The primary objective in the bacteriological examination of water is the detection of treatment deficiencies which can result in the entry of pathogens into the finished water supply. However, the occurrence and density of viral and bacterial pathogens in polluted waters is highly variable. This variability reflects the intestinal diseases that are prevalent at a given time in the human or other animal subpopulations which contribute their wastes to a particular effluent or watershed, as well as the numerous physical and chemical conditions which affect microbial virulence and survival.

The relative number of coliforms and viruses in feces, sewage, and polluted surface waters have been considered by some to be important in evaluating the waterborne disease potential and validity of the coliform standard (6). While coliforms are discharged in the feces of nearly everyone, viral excretion is generally limited to children under the age of 15 years with approximately 10% of these children shedding viruses at any given time. About 30% of the population falls into this age group, and calculations based on a *per capita* daily discharge of virus and coliforms result in estimated virus-coliform ratios of 1:65,000 for feces, 1:92,000 for sewage, and 1:50,000 in polluted surface water (1); studies made on Missouri River waters indicate a ratio of 1:500,000 (7). First year results of an in-house study (Brashear, unpublished) of polluted surface water have shown that even with improved viral methodology, no apparent relationship exists between the number of total or fecal coliforms in water and the number of viruses (Table 1). Viral isolates which included poliovirus 1,2,3, coxsackievirus B2, B4, and echovirus 1, 4, 25, were obtained from sample volumes of approximately 380 liters (100 gallons); virus to fecal coliform ratios ranged between 1:23 to 1:11,000. Although these ratios are considerably lower than previously reported, presumably due to improved recovery methodology, the coliform bacteria obviously greatly outnumber enteric viruses in sewage and polluted surface waters and thus still appear to be a more sensitive indicator of possible pathogen occurrences.

An important point which should be noted with respect to virus-coliform ratios is relative sample volumes. Traditionally, coliform densities in waters have been expressed as organisms per 100 ml or the approximate volume of a glass of water (Butterfield, personal communication). The standard drinking water sample volume for membrane filtration analysis was recently raised from 50 ml to 100 ml. By way of contrast, the methodology now being developed for isolating viruses from water (potable, surface, ground) processes from 1 to 1,900 liters, and typical finished water sample volumes range from 380–1,900 liters. For equivalent density comparisons between viruses and coliforms, it is necessary to analyze comparable water volumes of a liter or more for each microorganism.

Efficacy of Water Treatment Processes

A recent field study reported by Akin et al. (1) examined finished water quality of six municipal treatment plants in three states. The treatment plants use surface waters as their principal raw source water and rely on conventional treatment practices, i.e. flocculation, sedimentation, filtration, and chlorination (Table 2). Bacteriological quality of the source waters was variable, and at times exceeded the recommended maximum fecal coliform density (2000/100 ml) for surface water sources (19). Although the primary objective of this study was to determine whether human enteric viruses could be detected in finished waters using the equipment/procedures currently under development, parallel bacteriological analyses

TABLE 1. MICROBIOLOGICAL QUALITY OF OHIO RIVER WATER FROM CINCINNATI AREA*.

Location[1]	Total Coliforms (per 100 ml)	Fecal Coliforms (per 100 ml)	Standard Plate Count per ml	Virus[2] Isolates
Schmidt (467)[3]	11,000	6,500	37,000	0
Southside (475)[3]	49,000	17,000	62,000	3
Schmidt	85,000	23,000	79,000	9
Schmidt	38,000	11,000	48,000	1
Public Landing (470)[3]	3,400	370	20,000	16
Schmidt	105,000	43,000	162,000	17
Public Landing	35,000	3,500	112,000	1
Schmidt	99,000	42,000	193,000	31
Public Landing	48,000	7,200	46,000	12
Southside	135,000	33,000	79,000	59
Public Landing	7,300	1,200	8,300	3

* From Brashear

[1] Sampling period 9/8/75 through 12/1/75

[2] From 100 gallon (380 liters) water samples

[3] Ohio River Mile

TABLE 2. PHYSICAL/BACTERIOLOGICAL SOURCE WATER CHARACTERISTICS AT WATER TREATMENT PLANTS*.

Location	Source Water(s)	Capacity/mean Output (10,000 m^3/day)	Fecal Coliforms (MPN/100 ml)	F.C. Geometric Mean
Columbus, OH	Scioto River	26.5 / 13.6	38-3,000	280
Sidney, OH	Tawana Creek, wells	1.4 / 1.1	40-2,800	263
Muncie, IN	White River	6.0 / 4.9	50-2,000	210
Seymour, IN	E. Fork White River	0.56/ 0.45	11-14,000	536
Kansas City, MO	Missouri River	79.5 / 41.5	1,700-4,300	3,010
St. Joseph, MO	Missouri River	11 / 5.3	280-7,000	1,890

* From Akin *et al.* (1)

TABLE 3. TOTAL COLIFORM/FECAL COLIFORMS RESULTS OF FINISHED WATER SAMPLES FROM SIX TREATMENT PLANTS*.

Treatment Plant Location	Membrane Filter[a]		Modified MPN[b]		Large Volume Sampler[c]	
	Total Coliforms	Fecal Coliforms	Total Coliforms	Fecal Coliforms	Total Coliforms	Fecal Coliforms
Columbus, OH	0/4[d]	0/4	2/4	0/4	3/4	0/4
Sidney, OH	1/3	1/3	2/3	1/3	0/3	0/0
Muncie, IN	2/4	0/4	0/4	0/4	3/4	2/4
Seymour, IN	3/11	0/11	5/11	1/11	10/11	6/11
Kansas City, MO	0/2	0/2	2/2	0/0	0/0	0/0
St. Joseph, MO	1/1	0/1	0/1	0/1	—	—

* From Akin *et al.* (1)

[a] Up to 5 liters tested; negative results indicate a value <0.02/100 ml; the largest value obtained was 0.12 organisms/100 ml.

[b] 5.5 liters tested; negative results indicate a value <0.02/100 ml; the largest value obtained was 0.23 organisms/100 ml.

[c] 380 liters tested; negative results indicate a value <0.00059 organisms/100 ml; the largest value obtained was >0.0043/100 ml.

[d] Occurrences/Number of Samples

involving large volume sampling protocols (380 liters) were performed to establish coliform densities in finished waters.

A summary of the bacteriological findings is presented in Table 3. Using the membrane filter procedure with up to 5 liters of finished water, coliforms were detected in seven samples; one sample also contained fecal coliforms. The modified MPN procedure (5.5 liter samples) found coliforms in 11 samples, 2 of which also contained fecal coliforms. Finally, the large volume sampling method (380 liters) found coliforms in 17 samples, 8 of which also contained fecal coliforms. In all cases, coliform densities were below the maximum contaminant level (MCL) set by NIPDWR. More importantly, in 56 finished water samples in which the average volume of water analyzed was 1,100 liters, *no* viruses were isolated. Previous laboratory evaluations of the sensitivity of the virus recovery procedure indicated a detection level of 3-5 units (poliovirus) per 380 liters when 1,900 liters were sampled (14). In contrast, the large volume sampling protocol for coliforms was able to detect organisms using only 380 liter sample volumes.

Thus, the results of this study support the adequacy of conventional treatment processes for removal of bacterial and viral pathogens from even the poorest quality source waters (2,3,4). These data show that at least 99.9999% coliform reduction is readily attainable in water treatment facilities with complete treatment; this percent reduction is based on data in Table 3 where a negative large volume sampling result from a highly polluted source water indicates a coliform density of < 0.00059/100 ml. Filtration, particularly when preceded by chemical flocculation, is very effective in removing microorganisms (17). While the majority of water treatment facilities utilize rapid sand filtration prior to chlorination and distribution, a number of utilities lack filtration capabilities and rely solely on chemical disinfection to eliminate microbial pathogens. Such treatment deficiencies have been associated with increased outbreaks of giardiasis (8).

Epidemiological Findings

Twelve outbreaks of giardiasis including 4,800 cases in Rome, New York, were documented during the period 1971–1974 (8,9). all of these outbreaks, with the exception of Aspen, Colorado, involved municipal systems which used surface water supplies that were only chlorinated prior to distribution. *Giardia*, a flagellated protozoan, forms cysts which are not inactivated by chlorination at the dosages and contact times normally employed in water treatment processes; whether coliforms were present or absent from systems associated with the outbreaks has not been established. Although high chlorine concentrations and protracted contact periods will inactivate the cysts, it is recommended that filtration be implemented as part of the complete treatment of all surface waters. It is possible that with unfiltered surface waters, the coliform indicator system may not be adequate to signal the presence of *Giardia* or other pathogenic cyst forming protozoans. Since the analysis for this organism is time-intensive, complex, and imprecise, the logical approach is the elimination of the problem through improved water treatment practices, especially for finished water systems which have had a previous (chronic or intermittent) history of waterborne disease outbreaks attributed to treatment deficiencies. Regarding the etiology of waterborne outbreaks of gastroenteritis, hepatitis-A, shigellosis, typhoid, and salmonellosis, Craun *et al.* (9) concluded that treatment deficiencies, such as inadequate disinfection, interruption of disinfection or lack of treatment of ground waters were the primary causes.

Conclusions

With the implementation of the National Interim Primary Drinking Water Regulations, the coliform group of microorganisms remains the most relevant sanitary index for finished waters. Recent findings confirm earlier work that the coliform indicator concept is adequate in assuring bacterial and viral safety of water. The inability of present virological methodology to isolate viruses from finished waters results in continuation of our reliance on and confidence in the coliform group for monitoring finished water quality.

Although surface water quality may vary widely and must be continually improved through protection of receiving waterways, present water treatment practices are capable of consistently producing pathogen-free water to the consumer. The critical biological safeguards in water treatment continue to be flocculation/filtration with post-chlorination. Excluding protozoan cysts, all bacterial and viral pathogens are readily inactivated by chlorine dosage and contact times routinely used in properly operating water treatment facilities. The key to eliminating waterborne disease outbreaks, however, is constant surveillance of finished waters and their distribution network and the maximum use of good treatment processes. Treatment plants with any deficiencies must be upgraded to remedy these inadequacies and assure production of the best quality potable water.

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QUESTION AND ANSWER SESSION

G.W. Fuhs, Division of Laboratories & Research, New York Department of Health, Albany, New York

Is a standard plate count on groundwater more sensitive to contamination which may have occurred one week previous to sampling?

M.J. Allen, Water Supply Research Laboratory, U.S. Environmental Protection Agency, Cincinnati, Ohio

Waters from undisturbed or protected aquifers, unlike those from surface sources, have low indigenous bacterial populations. However, in shallow aquifers where percolating water from surface sources can provide sufficient nutrients to support microbial growth, an unusually high bacterial population could indicate infiltration of contaminated water into the aquifer. More importantly, abnormally high microbial levels in shallow aquifers can adversely affect detection of the more specific indicators. Thus, coliforms could be undetectable in ground water where excessive bacterial densities occur. The failure to detect coliform bacteria results, unfortunately, in the assumption that these contaminated supplies are safe to drink. Therefore, it could be necessary to employ additional microbial indicator groups for ground water. These indicator organisms may supplement or replace the coliform group when ground water conditions are such that erroneous results could occur.

Interferences to Coliform Detection in Potable Water Supplies

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The monitoring of potable water supplies must always utilize bacteriological procedures that are as sensitive for coliform detection as the state-of-the-art permits. This goal is of particular importance in a continuing surveillance for low levels of coliform bacteria that signal water treatment or transmission deficiencies which could allow the breakthrough of waterborne pathogens. While there has been much effort directed towards improving methodology, standardization of testing procedures, and achieving greater levels of data reliability through quality control and laboratory certification programs, little attention has been given to those peripheral water characteristics that may impact on the detection of indicator organisms. This concern has become more critical with the change of the official MF test volume from 50 ml to 100 ml in the Drinking Water Standards, in order to obtain better statistical reliability at the baseline limit of one coliform per 100 ml. These interferences relate to the nature and density of potable water turbidity or to the composition and magnitude of the finished water microbial flora.

Turbidity in Potable Water

Turbidity in potable water may be derived from source water, water treatment practices, problems in the distribution system, or a combination of these factors. Thus, turbidity may range from algal growth, natural siltation, decaying vegetation, industrial waste discharges to surface waters, chemicals in ground water, poor filtration practices, residual coagulants in water treatment, corrosion, microbial growth or soil intrusions in water distribution systems. As a result of these conditions, a variety of materials may be represented in turbidity, some of which can impart a chlorine demand. Other turbidities may be sources of interference to coliform detection, and only a few kinds of material will be of little concern as protective habitats for microbial survival.

Turbidity can be a transport vehicle for bacterial penetration of treatment barriers. While sand particles may be of little concern, other natural mineral turbidity, hydrated oxides from flocculation, and organic debris have been shown to provide absorption sites for bacteria (16,17,18,22). These conditions are of major concern in those supplies that employ disinfection as the sole treatment process for raw surface waters (22). Without use of more extensive treatment processes, the barrier to microbial penetration becomes inadequate because of turbidity fluctuations caused by stormwater runoff, algal blooms, seasonal turnover of impounded waters and water uses upstream. Where water treatment processes are poorly controlled, natural turbidity, alum floc particles or cationic polymers used in coagulation may break through filter beds into the distribution system. These conditions will produce turbidity levels higher than 1 NTU and may bring associated bacteria into the potable water.

Turbidity Interferences

Suspended material (turbidity) in a potable water sample may preclude use of the membrane filter coliform procedure. Reasons for rejecting the MF procedure relate to the volume of samples that can be filtered, character of suspended material, and the thickness of the suspended material that deposits on the membrane filter surface.

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Relatively thin layers of gelatinous, finely divided, or hygroscopic materials, such as suspended ferrous, manganese, and alum flocs or algal cell populations may clog filter pores or cause a confluent film of growth to develop during incubation. Thick surface layers of crystalline or siliceous materials may cause little or no difficulty.

There is no practical method of removing turbidity without also removing some unpredictable portion of the bacterial population including coliform bacteria in the sample. Our experience has indicated that pre-filtering turbid water through a coarse filter or decanting the supernatant from settled turbidity in a water sample results in a 20 to 80 percent loss of organisms. Turbidity limits for membrane filtration have never been determined because the limits vary with the type of material (sand vs. clay), chemical composition (phosphates, coagulants) and presence of various algal masses. While the multiple tube method may be the method of choice when testing potable water samples that contain turbidity interferences to membrane filtration, this approach may have some undesirable consequences. Selection of the multiple tube method automatically limits routine analyses to a 50 ml test portion, rather than the 100 ml test portion used with the membrane filter procedure. Thus, the theoretical detection limit for total coliforms in potable water becomes 2 organisms per 100 ml and assumes that these organisms are uniformly distributed in two 50 ml test portions. However, uniformity of bacterial suspension may not be achieved in turbid samples. Adsorption of bacteria to turbidity particles and settling of heavier particles are factors that must be rectified since the random occurrences of only three coliforms in the 50 ml test portion is sufficient to produce an unsatisfactory result. Coliforms trapped in turbidity particles may not produce gas in the presumptive medium unless released by vigorous sample agitation. This condition may be the cause of occasional differences in results from duplicate samples analyzed at the local water plant laboratory and at the State laboratory after indeterminant intervals of agitation during transit.

Microbial Interferences

While water produced at the treatment plant may be of high bacteriological quality, finished water entering a distribution network is subject to continuous bacteriological deterioration (1,4,7,8,10,12,19,20). Some factors which contribute to the deterioration include: 1) inadequate dosage or loss of disinfectant residual; 2) incomplete removal of organics and or turbidity due to treatment deficiencies; 3) seasonal temperature fluctuations; 4) extended retention periods in reservoirs, standpipes, and dead-end water mains; 5) sediment accumulations; and 6) line breaks and cross-connection occurrences in the distribution network. Although the microbiological flora in finished waters is highly variable, bacterial groups most frequently associated with finished water deterioration include: *Pseudomonas, Flavobacterium, Achromobacter, Proteus, Klebsiella, Bacillus, Serratia, Corynebacterium, Spirillum, Clostridium, Gallionella, Arthrobacter,* and *Leptothrix* (6). These organisms, in general, are able to survive and many may even multiply in finished waters. Such bacterial regrowth may result in slime deposits, tubercle formation, development of taste and odor problems, accelerated pipe deterioration (6,12,20,21) increased health risk (14) and may also interfere with bacteriological monitoring procedures.

Although finished waters which have been shown to be coliform free are generally considered safe for human consumption, there are occasions when the absence of detectable coliforms cannot be equated with potability. Excessive densities of non-coliform organisms in finished water may desensitize assay procedures for total coliforms (5,9,11,23). These non-coliform organisms can coexist with coliforms in water, but when introduced into lactose broth, many of the non-coliform organisms may multiply at a rapid rate, thus intensifying the factor of coliform inhibition (15) or simply outgrow a smaller coliform population.

In laboratory tests conducted by Hutchison et al (17), suspensions of various antagonistic organisms *(Pseudomonas, Actinomyces, Sarcina,* and *Micrococcus)* in a density range of 1,000 to 2,000 per ml, when added to lactose tubes simultaneously to a suspension containing 10 *E. coli* per ml, resulted in reduced coliform detection. The loss of sensitivity ranged from 28 to 97 percent, depending upon the combination of mixed strains in these experiments. Considering that the baseline for coliform detection is one organism per 100 ml, any loss of test sensitivity becomes critical.

Analysis of bacteriological data from survey of distribution water samples from 969 public water plants in this nation suggested that interference with coliform detection may be a problem (13). These data (Table 1) showed that the frequency of detecting total and fecal coliforms by the MF procedure, increased as the standard plate count increased to levels up to 500 per ml, but decreased in frequency when the non-coliform bacterial population exceeded 1,000 organisms per ml. These findings again suggest that high densities of non-coliform bacterial populations have an adverse affect on detection of the coliform indicator.

While provisions for a Standard Plate Count measurement are not currently included in the National Interim Primary Drinking Water Regulations, field studies in several different geographical areas are now in progress to obtain additional information on the impact of the total microbial populations on MPN and MF methodology. One of these studies involves the Cincinnati water distribution system. This public water supply network, like many other systems, has numerous dead-end water mains. The water in these low flow mains is subject to abnormal deterioration, as demonstrated by the number of customer complaints in the areas of a number of these low flow sections. For this reason, the Water Distribution Division-Maintenance Section of the Cincinnati Water Works has maintained a routine flushing program for at least 30 years.

Weekly, a number of troublesome dead-end water mains are flushed to clear out accumulated sediments and bring fresher waters with free chlorine residuals into these mains. During a two year period, with the cooperation of the Maintenance Section personnel, samples from these flushes were obtained for bacteriological analysis. All samples were iced immediately and processed within five hours of collection. Bacteriological analyses of 613 water samples from 32 sampling sites included the total coliform multiple tube and membrane filter procedures and a

TABLE 1. BACTERIAL PLATE COUNT VS. COLIFORM DETECTION IN DISTRIBUTION WATER NETWORKS FOR 969 PUBLIC WATER SUPPLIES.

General Bacterial Population*		Total Coliform		Fecal Coliform	
Density Range per 1 ml	Number of Samples	Occurrences	Percent	Occurrences	Percent
<1 - 10	1013	47	4.6	22	2.2
11 - 30	371	28	7.5	12	3.2
31 - 100	396	72	18.2	28	7.1
101 - 300	272	48	17.6	20	7.4
301 - 500	120	30	25.0	11	9.2
501 - 1,000	110	21	19.1	9	8.2
>1,000	164	31	18.9	5	3.0
TOTAL	2446	277	—	107	—

* Standard Plate Count (48 hrs. incubation, 35° C)

standard plate count (SPC agar pour plates, incubated at 35° C for 48 hours). Physical/chemical parameters measured were free chlorine residual (DPD test), turbidity, water temperature and pH.

Bacteriological analysis of water samples obtained from dead-end water mains (Table 2) showed that 76 of 613 samples contained coliforms when examined by the MPN procedure. In contrast, the membrane filtration technique detected coliforms in only 19 of these samples. Thus, the MF method detected coliforms in only 25% of the samples that were positive for coliforms by the MPN method. Closer examination of the data shows a correlation between excessive bacterial densities and desensitization of the membrane filter procedure.

While detection of coliforms by both the MPN and MF techniques increased as the standard plate count increased, the MF method detected disproportionately fewer coliforms as noted in the range of MF coliform values compared to the MPN coliform values per 100 ml. Although it is not apparent from the data in Table 2 that the MPN method is also subject to interferences by SPC organisms, a limited number of water samples examined by the method displayed growth inconsistencies. Several MPN tubes that showed no gas, but heavy growth, were inoculated into brilliant green tubes and incubated at 35° C for 48 hours. These tubes did produce gas, indicating that the competing non-coliform bacteria had suppressed the normal metabolic pathway to gas production by coliforms in the presumptive medium. Such false-negatives were also noted with the MF procedure. Membrane filter cultures with excessive densities of colonies but no differentiated sheen colonies, were aseptically transferred into lauryl tryptose broth for evidence of gas production within 48 hours at 35° C then confirmed in brilliant green bile broth. This extension of the MF procedure also revealed the presence of undetected coliforms that were marked by high densities of the Standard Plate Count.

Controlling Interferences Through Treatment

Improved water treatment practice is the first line of defense against problems related to turbidity in finished water. With reduction of turbidity to below 1 NTU,

TABLE 2. COLIFORM OCCURRENCES VS. STANDARD PLATE COUNT.

	Standard Plate Count Range (per ml)		
	< 500	501-1000	> 1000
All Samples*	502	35	51
Samples with Coliforms** (MPN Method)	50	8	15
Percent of Samples	10%	22.9%	29.4%
MPN Range (per 100 ml)	2-49	5-79	2-920
MPN Mean (per 100 ml)	16	18	110
Samples with Coliforms*** (MF Method)	11	2	5
Percent of Samples	2.2%	5.7%	9.8%
MF Range (per 100 ml)	< 1-18	< 1-50	< 1-90
MF Mean (per 100 ml)	2	7	13
MPN % Occurrence less MF % Occurrence	7.8	17.2	19.6

* 588 of 613 samples had countable SPC

** 3 MPN positive samples did not have countable SPC

*** 1 MF positive samples did not have countable SPC

TABLE 3. MPN COLIFORM OCCURENCES VS. FREE CHLORINE RESIDUALS (DPD)*

Free Chlorine Residual (mg/l)	Number of Coliform Occurrences	SPC Average (bacteria/ml)
<0.1	58	3040
0.1-0.5	13	860
0.6-1.0	4	50
>1.0	1	79

* Typical values for finished waters were 1-2 mg/l free chlorine and Standard Plate Count < 1/ml.

disinfection efficiency increases and enhances control of bacterial regrowth throughout the distribution network. Analysis of the effectiveness of free chlorine residuals in controlling bacterial deterioration, as demonstrated in this study (Table 3) and in the Community Water Supply Study (Table 4) prove the feasibility of this approach for minimizing the potential non-coliform interferences to coliform methodology (6,8,22,23). Pipe sediment accumulation in low-flow water mains and dead-end lines is a source of both intermittent turbidity and nutritive materials that support bacterial persistance and possible regrowth problems that may cause coliform interferences. The total elimination of dead-end water mains is, of course, impossible, but implementation of practices such as periodic flushings, main cleaning, maintenance of *free* chlorine residuals and modification or addition of various treatment processes can substantially improve distribution water quality in dead-end mains and low flow sections.

Chemical characterization of 5 sediment samples from the Cincinnati distribution system confirms our argument that this material serves as a nutrient supply for bacterial regrowth. Dried sediment samples were pulverized with 0.10 gm of each

TABLE 4. THE EFFECT OF VARYING LEVELS OF RESIDUAL CHLORINE ON THE TOTAL PLATE COUNT IN POTABLE WATER DISTRIBUTION SYSTEMS*.

Standard Plate Count**	Residual Chlorine (mg/l)							
	0.0	0.01	0.1	0.2	0.3	0.4	0.5	0.6
< 1	8.1%	14.6%	19.7%	12.8%	16.4%	17.9%	4.5%	17.9%
1 - 10	20.4	29.2	38.2	48.9	45.5	51.3	59.1	42.9
11 - 100	37.3	33.7	28.9	26.6	23.6	23.1	31.8	28.6
101 - 500	18.6	11.2	7.9	9.6	12.7	5.1	4.5	10.7
501 - 1000	5.6	6.7	1.3	2.1	1.8	0	0	0
> 1000	10.0	4.5	3.9	0	0	2.6	0	0
Number of Samples	520	89	76	94	55	39	22	28

* Data from a survey of community water supply systems in 9 metropolitan areas (54)
** Standard Plate Count (48 hrs. incubation, 35° C)

sample added to 200 ml of distilled-deionized water and allowed to solubilize for 3 hours at ambient temperature. An aliquot of the supernatant was removed, acidified with HCl and analyzed with Dohrmann DC-52 carbon analyzer. Additional aliquots of the supernatant were removed for Kjeldahl-nitrogen and nitrate-nitrogen analysis. Kjeldahl-N ranged from 250 to 575 mg/l, nitrate-nitrogen from 4400-5200 mg/l and total organic carbon from 200 to 550 mg/l. Bacterial regrowth becomes a reality in slow flow sections or dead end lines where such high levels of bacterial nutrients occur in pipe sediments especially with water temperatures above 13° C, pH below 10, and no free chlorine residual or ineffective chloramines present. Laboratory growth studies using distilled water extracts of pipe sediments showed that nutrient concentrations were capable of supporting bacterial growth from initial densities of less than 100 SPC organisms per ml to 300,000 organisms per ml within 10 days at 20° C.

Controlling Interferences Through Methodology

Until all interferences created by inadequate water treatment and distribution practices are eliminated in problem water supplies, the laboratory must remain vigilant to the problem of coliform detection in potable waters of varying qualities. Interferences to coliform detection often go undetected, partly because of a Standard Methods concept that implies that no coliforms are present in a multiple tube test when there is no evidence of gas production within 48 hours, or with the membrane filter procedure when sheen colonies are absent. In the multiple tube procedure, consideration should be given to confirmation of test samples that produce 5 presumptive tubes with heavy growth but lack visible gas production. For membrane filter cultures that lack differentiated sheen colonies and that have high background densities of non-differentiated colonies, the entire membrane filter should be placed in lauryl tryptose broth to test for gas production followed by confirmation in brilliant green bile broth. This procedure would yield qualitative information on suppressed coliform occurrences that might otherwise go undetected. Since some types of turbidities between 1 and 5 NTU physically block membrane filter pores, impede flow through and prevent development of discrete differential colonies, the laboratory will be obligated to use the multiple tube procedure until the interference problem is corrected through improved treatment or distribution system maintenance.

Conclusion

In an effort to detect coliform occurrences at a baseline of one organism per 100 ml, potable water must be free of interferences from turbidities that exceed the 1 NTU limit specified in the Interim Primary Drinking Water Regulations and bacterial densities over 500 organisms per ml should not be tolerated. To some extent, these interferences may be circumvented in the laboratory through alterations in methodology. However, these measures should only be considered temporary expedients to the monitoring program. The ultimate solution to these issues resides in improved treatment practices and attention to minimizing deterioration of water quality during distribution.

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QUESTION AND ANSWER SESSION

C.D. Cameron, Fairfax County Water Authority, Occoquan, Virgina

What effect would public announcements urging residents to use less water during water shortages have upon the distribution system water quality? Would not a long holding time (2 weeks versus 2 days) in system mains and tanks encourage loss of Cl_2 and an increase in total population?

E.E. Geldreich, Water Supply Research Laboratory/MERL, U.S. Environmental Protection Agency, Cincinnati, Ohio

There are several factors not defined that will impact on the status of the standard plate count population in a distribution network during slow flow periods. During periods of warm water temperature (above 13° C) and increased chlorine demand in the distribution lines, regrowth of the bacterial population will occur. Some of this regrowth potential might be suppressed where the finished water pH ranges from 9.0 to 10.0. If chloramines are carried as a residual, longer contact time in the distribution network would result in some limited control over bacterial regrowth. As a temporary counterforce, less abrupt changes in water pressure during this period would reduce the mixing of sediments into the water flow. However, our data would suggest the overall effect would be a gradual degradation of the water regardless of limited benefits achieved by longer contact time and water quiescence.

R.R. Colwell, Department of Microbiology, University of Maryland, College Park, Maryland

Since 90% of the total count is missed when pour plates of water samples are incubated at 35° C, why not use lower incubation temperatures?

E.E. Geldreich

We recognize that not all viable bacteria from a mixed population in a water sample can be cultivated on a single medium, at any one incubation temperature or for one specified incubation time. The critical concern with the general population of bacteria in potable water lies primarily in the potential interferences they may create in the test for coliforms. The other determining factor is to attempt to include some indirect measurement for *Pseudomonas*, *Flavobacterium*, and other secondary pathogenic invaders which could pose a health risk in the hospital environment. Since the coliform test is incubated at 35° C and the potential secondary pathogenic invaders optimally grow at body temperature, the choice of a non-specific bacterial count in water at 35° C was logical. Furthermore, any attempt to specify a limit for this portion of the general bacterial population must follow strict adherence to a specific protocol for medium, incubation temperature and time. Thus the expression "Standard Plate Count" becomes appropriate and the bacterial population limited to growth on Standard Plate Count agar, with incubation of pour plate cultures at 35° C for 48 hours.

Impact of the Coliform Standard on the Transmission of Disease

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Dr. Hendricks, the Symposium Chairman, has asked that I review and summarize the data on waterborne disease outbreaks in the United States, concentrating on the water system deficiencies that allowed the outbreaks to occur. I will also discuss the bacteriological quality of water provided by the water supplies involved in outbreaks and whether bacteriological surveillance as proposed in the National Interim Primary Drinking Water Regulations will prevent the occurrence of waterborne outbreaks.

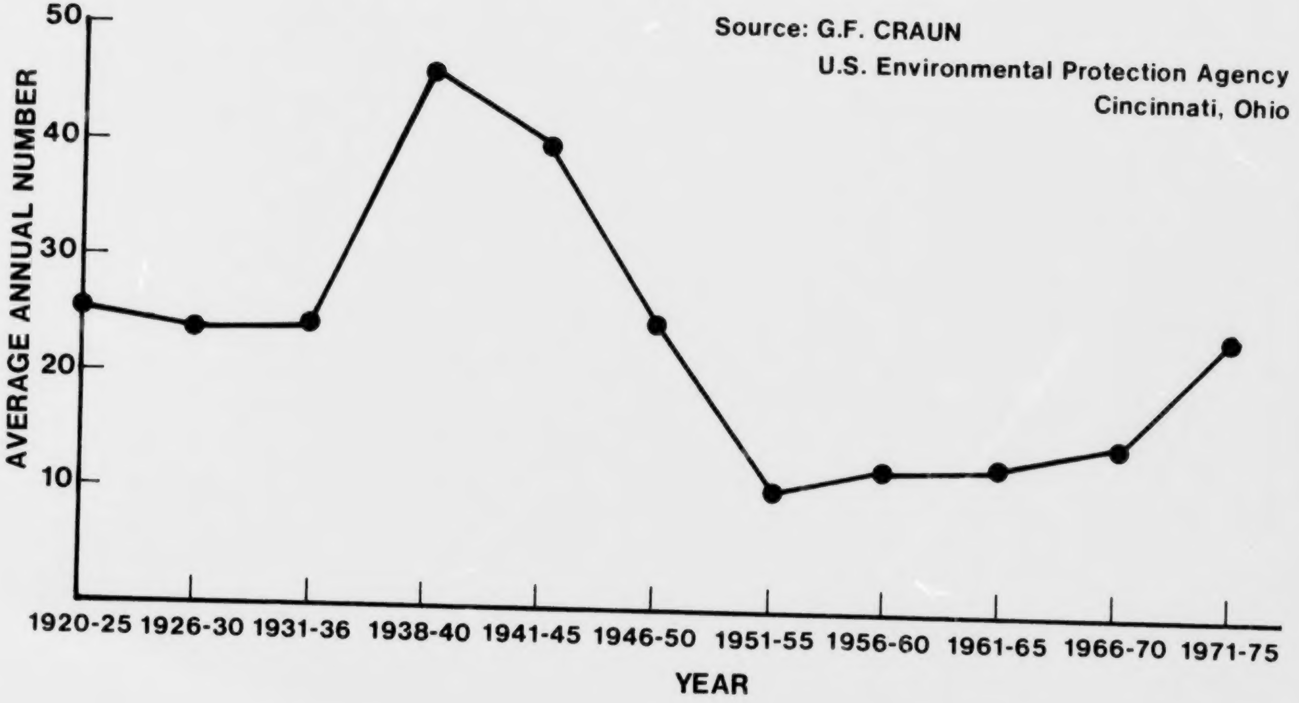
The Center for Disease Control (CDC) and the Environmental Protection Agency (EPA) jointly conduct surveillance of waterborne disease. Outbreaks are reported to the CDC and the EPA by state and local health departments and by water supply agencies. This report summarizes data reported during 1971-1975.

Only outbreaks associated with water used for drinking or domestic purposes are included in this analysis. To be considered an outbreak at least two cases of infectious disease must be reported before a common source can be noted and investigated. Except in unique circumstances, such as a case of chemical poisoning in which the chemical was identified in the water, a single case cannot be recognized as having been caused by drinking water. The waterborne outbreaks reported here are those in which drinking water has been implicated epidemiologically as the vehicle of transmission of the illness. In most of the outbreaks, the water was found to be bacteriologically or chemically contaminated.

For analysis, the water systems were classified as municipal, semi-public, or individual. Municipal water systems are defined as public or investor-owned water supplies that serve communities. Individual water systems are those used exclusively by single residences in areas without municipal systems or by persons travelling outside of populated areas (e.g., backpackers). Semi-public water systems, located in areas not served by municipal systems, are developed and maintained for a group of residences (e.g., subdivisions and trailer parks) or at locations where the general public has access to drinking water (e.g., industries, camps, parks, resorts, institutions, and hotels). The definition of municipal water system and semi-public water system correspond to the Safe Drinking Water Act's definitions of community water system and non-community water system. The major exception is that small subdivisions and trailer parks are included as semi-public systems in this analysis, whereas, the Safe Drinking Water Act defines these as community water systems.

During the period 1971-1975, 123 waterborne outbreaks were documented in the U.S., resulting in almost 28,000 cases of illness (Table 1). The three largest outbreaks occurred in municipal water systems: Sewickley, Pennsylvania (5,000 cases of gastroenteritis) in 1975; Rome, New York (4,800 cases of giardiasis) in 1974; and Pico Rivera, California (3,500 cases of gastroenteritis) in 1971. Two deaths were associated with outbreaks of waterborne disease during this period.

The mean annual number of reported waterborne outbreaks during 1971-1975 was 25. This is twice as many outbreaks as the mean annual number reported during the period 1951-1970 and equals the mean annual number reported during the period 1920-1936 (Figure 1). The reason for this apparent increase in number of outbreaks is difficult to ascertain but is felt to be primarily the result of increased reporting and surveillance activities.



Source: G.F. CRAUN
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Figure 1. AVERAGE ANNUAL NUMBER WATERBORNE OUTBREAKS, 1920 - 1975.

TABLE 1. WATERBORNE DISEASE OUTBREAKS.
1971-1975

	1971	1972	1973	1974	1975	TOTAL
Outbreaks	19	29	26	25	24	123
Cases of Illness	5182	1638	1774	8356	10,879	27,829

The 123 waterborne outbreaks were classified by type of water system (Table 2). More outbreaks occurred in semi-public water systems (57%) than municipal systems (30%) and individual systems (13%); however, outbreaks in municipal systems affected an average of 504 persons compared to 129 persons per outbreak in semi-public and 9 persons per outbreak in individual systems. Although most of the outbreaks (57%) occurred in semi-public water systems, most of the illness (67%) resulted from outbreaks in municipal systems.

TABLE 2. WATERBORNE OUTBREAKS BY TYPE OF SYSTEM.
1971–1975

	OUTBREAKS	CASES OF ILLNESS
Municipal Systems	37	18,633
Semi-Public Systems	70	9,058
Individual Systems	16	138
Total	123	27,829

An etiologic agent was determined in only 49% of the outbreaks during the period 1971–1975 (Table 3). The remainder were characterized as acute gastrointestinal illness of unknown etiology. Outbreaks in which ill persons had symptoms including abdominal cramps, nausea, vomiting, and diarrhea 24 to 48 hours after consumption of water and in which no specific etiologic agent could be determined were grouped in this entity. This includes a clinical entity known as "sewage poisoning" which is presumably caused by either coliform organisms or certain viruses that have yet to be fully characterized.

TABLE 3. ETIOLOGY OF WATERBORNE OUTBREAKS.
1971–1975

	OUTBREAKS	CASES OF ILLNESS
Acute Gastrointestinal Illness	63	17,752
Hepatitis-A	14	368
Shigellosis	14	2,803
Giardiasis	13	5,136
Chemical Poisoning	12	511
Typhoid	4	222
Salmonellosis	2	37
Enterotoxigenic *E. coli*	1	1,000
Total	123	27,829

The most commonly identified pathogen during this period was *Shigella*; there were 14 outbreaks and 2,803 cases of shigellosis. Most of these occurred in non-municipal systems.

Giardia lamblia is a flagellated protozoan responsible for giardiasis. Clinical manifestations of *Giardia* infection can range from asymptomatic cyst passage to severe malabsorption. The mean duraction of illness is often 2 to 3 months. Only recently has *G. lamblia* been recognized as a cause of acute illness and as the etiologic agent in common-source outbreaks (3,2). The disease has received considerable attention since 1970, as a result of studies implicating tap water as the probable mode of transmission of giardiasis in travelers returning from Leningrad. This increased awareness of physicians and public health workers has no doubt been partially responsible for the increased reporting of this disease in the U.S. Fifteen waterborne outbreaks of giardiasis have been documented in the U.S. since the outbreak at Aspen, Colorado in 1965–1966. Thirteen occurred during the period 1971–1975, affecting 5,136 individuals.

Until the 1974 outbreak at Rome, New York, where 4,800 persons were affected, the waterborne outbreaks of giardiasis have generally involved small municipal systems or semi-public systems in recreational areas. Almost all of the outbreaks have occurred as the result of drinking untreated surface water or surface water whose only treatment was disinfection. *Giardia* cysts are not destroyed by chlorination at dosages and contact times normally employed in water treatment, but it is felt that they can be removed by well operated conventional treatment plants employing coagulation/flocculation, settling, and filtration.

Although adequate disinfection data are not currently available, it is felt that *Giardia* cysts are as resistant to chlorination as cysts of *E. histolytica*. If this is correct, high concentrations of chlorine and long contact times are required in situations where settling and filtration cannot be employed; this may necessitate dechlorination prior to distribution (1). It has been reported that *G. lamblia* cysts can survive in water and remain infective for sixteen days (5). Rendtorff (6) conducted a series of studies with prisoner volunteers and found that 8 of 22 men who received 10–25 *G. lamblia* cysts became infected and all 13 men who received from 100–1,000,000 cysts became infected. Because an obvious source of gross human contamination could not be identified in most of the waters systems having giardiasis outbreaks, some wild and domestic animals are suspected as being important in the transmission of the disease to man. It is possible in water systems using surface water whose only treatment is chlorination at conventional dosages and contact times that sufficient disinfection will be employed to produce a coliform-free water but not a giardia-free water.

The were 14 outbreaks of waterborne viral hepatitis affecting 368 people during 1971–1975. There has been considerable controversy regarding the existence of viruses in treated water supplies and the possible health consequences. Hepatitis-A has been epidemiologically implicated in 66 waterborne outbreaks since 1946 and the data can be examined to determine how these viral outbreaks occurred. Of the 22 outbreaks occurring in municipal systems, three resulted from either inadequate or interrupted disinfection and five were related to the use of contaminated, untreated surface or ground water. Half (eleven) of the outbreaks in municipal systems occurred as the result of contamination of the distribution system, primarily through cross-connections and backsiphonage. There is no evidence from waterborne outbreak data that hepatitis-A virus has been transmitted through water systems with properly operated conventional treatment plants except where distribution system deficiencies have been found as the source of contamination.

There were 12 chemical poisonings involving fluoride, chromate, selenium, phenol, furadan, arsenic, ethyl acrylate, a mixture of lubricating oil and kerosene, cutting oil, fuel oil, and a herbicide.

The four typhoid outbreaks affected 222 people and involved semi-public and individual water systems. The largest outbreak, responsible for 210 cases of typhoid,

TABLE 4. WATERBORNE DISEASE OUTBREAKS BY TYPE OF DEFICIENCY
1971-1975

	OUTBREAKS	CASES OF ILLNESS
Untreated Surface Water*	19	5,729
Untreated Ground Water	38	3,958
Treatment Deficiencies	39	10,139
Distribution System Deficiencies	15	7,468
Miscellaneous	12	535
Total	123	27,829

* Includes seven outbreaks of giardiasis in which surface water was chlorinated but not filtered

occurred in February and March 1973, at the South Dade Migrant Farm Labor Camp, Dade County, Florida. This was the largest reported outbreak of typhoid fever in the U.S. since 1939 (4). No deaths occurred, and the outbreak subsided with few secondary cases and no transmission to persons not connected with the camp.

Two small outbreaks of salmonellosis occurred during this period: 34 cases of *S. typhimurium* in a semi-public water system and 3 cases of salmonellosis in an individual water system.

At Crater Lake National Park in 1975, an estimated 1,000 cases of diarrhea occurred following contamination of a spring, which served as the drinking water source for the park, by overflow from an obstructed sewerage line. Enterotoxigenic *E. coli* serotype 06:H16 were isolated from ill persons and from water samples.

Untreated or inadequately treated water was responsible for the majority of waterborne outbreaks during 1971–1975 (Table 4). Treatment deficiencies, primarily inadequate or interruption of chlorination, and the use of untreated, contaminated ground water accounted for 63% of all outbreaks and 51% of all illness.

The outbreaks and cases of illness were also classified by cause and type of water system (Tables 5-6). As in the previous 25-year period, the major cause of outbreaks in municipal water systems during 1971–1975 was contamination of the distribution system; 41% of the outbreaks in municipal water systems occurred because of deficiencies in the distribution of water. Generally, contamination of the distribution system has occurred through cross-connections and backsiphonage, and the resulting outbreaks have been quite contained, affecting relatively few people. However during 1971–1975, outbreaks caused by contamination of the distribution system were responsible for 40% of the total cases of illness occurring in municipal water systems. Two large outbreaks accounted for most of the illness in this particular category: an estimated 5,000 cases of acute gastroenteritis in Sewickley, Pennsylvania, which was felt to be related to contamination of an uncovered storage reservoir for treated water and 1,400 cases of a similar illness in Sellersburg, Indiana, which was traced to sewage contamination of a water main during construction.

In the previous 25-year period, use of untreated ground water was responsible for most illness in municipal water systems. During 1971–1975, four outbreaks occurred in municipal systems because of the use of untreated ground water; however, only 74 (0.4%) cases of illness resulted, because the outbreaks occurred in small water systems. Nine (24%) outbreaks and 5,322 (29%) cases of illness occurring in municipal water systems were related to treatment deficiencies; six (16%) outbreaks

TABLE 5. MUNICIPAL WATER SYSTEMS DEFICIENCIES ASSOCIATED WITH WATERBORNE OUTBREAKS.
1971-1975

	OUTBREAKS	CASES OF ILLNESS
Untreated Surface Water*	6	5,404
Untreated Ground Water	4	74
Treatment Deficiencies	9	5,322
Distribution Deficiencies	15	7,468
Miscellaneous	3	365
Total	37	18,633

* Includes four outbreaks of giardiasis in which surface water was chlorinated but not filtered

and 5,404 (29%) cases of illness to use of untreated surface water. Four of the five giardiasis outbreaks occurring in municipal water systems were related to the use of surface water where disinfection was the only treatment. These were included in the "untreated surface water" category rather than the "treatment deficiency" category because specific treatment was not provided to remove or inactivate this pathogen. Conventional disinfection as the only treatment of surface water would not be expected to destroy *Giardia* cysts.

Use of untreated ground water was responsible for most of the outbreaks and cases of illness that occurred in semi-public water systems in the past. Although this is still an important problem, the 1971-1975 data show that deficiencies in treatment were also responsible for many outbreaks and illness occurring in these water systems. Use of untreated ground water and treatment deficiencies accounted for 81% of the

TABLE 6. SEMI-PUBLIC WATER SYSTEMS DEFICIENCIES ASSOCIATED WITH WATERBORNE OUTBREAKS.
1971-1975

	OUTBREAKS	CASES OF ILLNESS
Untreated Surface Water*	7	237
Untreated Ground Water	27	3,859
Treatment Deficiencies	30	4,817
Distribution Deficiencies	0	0
Miscellaneous	6	145
Total	70	9,058

* Includes three outbreaks of giardiasis in which surface water was chlorinated but not filtered

TABLE 7. SEASONAL DISTRIBUTION OF WATERBORNE OUTBREAKS, 1971–1975

	MUNICIPAL	SEMI-PUBLIC	
		TOTAL	AFFECTING VISITORS
January	1	2	1
February	2	1	0
March	3	2	1
April	2	4	4
May	1	9	6
June	5	14	12
July	4	12	10
August	5	12	11
September	4	4	4
October	3	3	1
November	4	3	1
December	2	4	3
Unknown	1		
Total	37	70	54

outbreaks and 96% of the cases of illness in semi-public systems. This is most likely related to the size and nature of these systems, since most semi-public systems would use ground water. Ground water is generally thought to be free of contamination and little may be done to prevent intermittent contamination of these sources. Where disinfection is provided, for either contaminated sources or as a preventative measure for intermittent contamination, little or nothing may be done to insure that continuous and effective disinfection is maintained. Many of these systems may be operated for only a part of the year, and this may explain the lack of proper operation and maintenance.

A large number of waterborne outbreaks each year involves the travelling public (Table 7). In 1971–1975, 54 of the 70 outbreaks (77%) in semi-public water systems affected travelers, campers, visitors to recreational areas or restaurant patrons. Thirty-nine of the 54 outbreaks (72%) involving a transient population occurred during the months of May-August, the period when outdoor activities such as picnicking, camping and vacationing are most common. This implies that there is either increased contamination of these water supplies during this period or, if it is assumed that the supplies are always contaminated, use by greater numbers of susceptible individuals during this period. Outbreaks in semi-public water systems peaked during the summer months in 1971–1975, which is consistent with the previous 25 years of record. There appears to be little seasonal variation for outbreaks in municipal water systems, either during this period or the previous 25-year period.

Waterborne disease surveillance such as this is important for several reasons. First and most important, it helps stimulate the recognition and investigation of ongoing outbreaks and terminate them by identification of the water supply deficiencies that allowed the outbreaks to occur. Second, it identifies deficiencies in water treatment practices and leads to the formation of control measures. Regulatory agencies can use the data for establishing water supply surveillance priorities based on outbreak-related problems. For example, these data would indicate that additional emphasis should be placed on surveillance of smaller water supplies, particularly the semi-public water supplies that serve the travelling public. For these systems, increased emphasis should be placed on protection of the source water from contamination, disinfection of these water supplies, and proper operation of the systems to insure that continuous, effective disinfection is maintained. Outbreaks in municipal systems would indicate that additional emphasis is needed in the area of cross-connection control and prevention of distribution system contamination. Third, waterborne disease surveillance can assist in the recognition of important or new etiologic agents so that appropriate water treatment can be provided. For example, the data indicate that conventional disinfection as the only treatment for surface water sources is ineffective in preventing waterborne transmission of giardiasis and that to protect against transmission of this disease all surface water should be treated by sedimentation and filtration in addition to chlorination.

TABLE 8. SUMMARY OF BACTERIOLOGICAL DATA FROM WATER SYSTEMS WHERE LOW NUMBERS OF TOTAL COLIFORMS WERE FOUND IN TAP WATER SAMPLES DURING THE OUTBREAK INVESTIGATION.

CAUSE OF OUTBREAK:

I. USE OF UNTREATED WATER
1. 1/100 ml, 21/100 ml, TNTC
*2. (-), 3/100 ml, 3/100 ml, 8/100 ml, 19/100 ml, 32/100 ml
**3. (-), (-), 200/100 ml
4. (-), 2/100 ml, 3/100 ml, 6/100 ml, 10/100 ml, 13/100 ml
5. (-), (-), (-), 162/100 ml

II. TREATMENT DEFICIENCY
1. (-), (-), 8/100 ml, 20/100 ml
†2. 15/100 ml
††3. <2.2 MPN
†††4. 12 samples all (-)
5. (-), 7200/100 ml
††††6. 4 samples (-), 3/100, 10/100 ml

III. DISTRIBUTION DEFICIENCY
1. (-)
2. (-)'s and low levels within DWS limits
3. 44 samples (-), 5/100, 8/100, 9/100, 10/100, 10/100, 26/100, >39/100, >39/100, 48/100, 52/100

IV. MISCELLANEOUS OR UNDETERMINED
1. 12/100, 22/100
2. (-)
3. 1/100, 1/100, 3/100, 4/100

Raw water data (coliforms) for above; not necessarily collected at the same time

*	246/100 ml
**	100/100 ml
†	29/100 ml
††	>16 MPN
†††	>50/100 ml
††††	>50/100 ml

Historical water quality data from water systems having a waterborne outbreak can be used to determine the effectiveness of bacteriological surveillance. Outbreaks during 1971–1975 with sufficient bacteriological data were examined to determine (1) the level of contamination at the time of the outbreak and (2) if routine bacteriological surveillance could indicate if an outbreak would occur.

In the majority of instances water samples collected during the outbreak investigation showed high levels of either total or fecal coliforms; however, there were outbreaks in which no or low numbers of coliforms were found. These were classfied according to the cause of the outbreak (Table 8). It is difficult to judge if these sample results are representative of the extent of contamination and illness occurred in the presence of low coliform contamination, or if the contamination was actually larger and the samples were just not collected at the right place or time.

There were five outbreaks in which the drinking water was epidemiologically implicated, but samples collected during the outbreak investigation were all negative for coliforms. One was caused by a cross-connection, one by contamination of the distribution system, and another by contaminated ice. It is felt that these samples were most likely collected after the contamination had passed through the system and are probably not representative of the water or ice which caused the illness. They were, however, representative of samples that might be collected for rountine bacteriological surveillance, and as such, the results would certainly be misleading. In the remaining two outbreaks with distribution samples negative for coliforms, contaminated water was distributed because of an interruption of treatment. This is known only because the treatment deficiency was documented and the untreated water was shown to be contaminated during the investigation. Unless distribution samples were collected at the time of the treatment failure, routine bacteriological surveillance would not have warned of a pending outbreak. There were several outbreaks where negative coliform samples were collected at the same time other samples were positive for coliforms. These examples serve to illustrate the importance of collecting timely samples and collecting samples at proper points in the system if a routine bacteriological sampling program is to be effective. Both timely and representative sampling are a function of the minimum number of samples required, and certainly a minimum of one or two samples per month or per quarter as permitted in the National Interim Primary Drinking Water Regulations (7) cannot properly assess the quality of water being consumed. In several outbreaks, it required two or more samples to find the contamination and in one, 44 of 54 samples collected were negative for coliforms. Unless samples can be collected continuously at a large number of points throughout the system, it is unlikely that routine bacteriological surveillance will be useful in preventing the occurrence of outbreaks. Since it is technically and financially infeasible to provide continuous bacteriological monitoring, alternatives must be considered if outbreaks are to be prevented. Monitoring for chlorine residual is a reasonable alternative and is especially important for small systems where relatively few bacteriological samples are required.

Outbreaks having routine bacteriological surveillance data were examined to determine if the occurrence of the outbreak could be predicted for that water supply. These were categorized by the cause of the outbreak to illustrate several points (Tables 9-12).

Previous bacteriological results were not available for those outbreaks caused by distribution deficiencies, but several observations can be made about routine bacteriological surveillance for prevention of these outbreaks. There are numerous examples of chemical cross-connections that have not resulted in illness, but this is because the water had a distinct color or odor not because of any routine surveillance. Bacteriologically contaminated water would not necessarily exhibit these characteristics, and it is highly unlikely that samples could be collected frequently enough to detect cross-connections and prevent outbreaks due to this cause. Several outbreaks occurred during 1971–1975 where bacteriological contamination entered the

TABLE 9. TWO OUTBREAKS CAUSED BY USE OF UNTREATED, CONTAMINATED WATER.

Outbreak in Upstate New York Restaurant
1971

Acute Gastrointestinal Illness
Overall attack rate 500-1000/581-1163 (86%)

Tap water samples collected in restaurant before and after outbreak were negative for coliforms except for one sample collected on 11/19/71 which showed 200 total coliforms per 100 ml. Spring on several occasions prior to the outbreak yielded 100 coliforms per 100 ml.

Big Sky Montana Outbreak
1975

Acute Gastrointestinal Illness (*Yersinia enterocolitica* was isolated from the water, but stools were not examined for this organism)
Overall attack rate 139/314 (44%)

It was reported that between 3/72 and 11/18/74 samples were collected every 1-2 months and that all had been negative for coliforms. Water samples obtained on 1/14-16/75 yielded from 1 coliform to >16 coliforms/200 ml. During the investigation of the outbreak samples from the two wells showed:

1. 4 samples - Negative for coliforms
2. 8 samples - Positive for coliforms

4 - 1/250 ml
2 - 2/300 ml
1 - 2/300 ml
1 - 7/250 ml

distribution system and routine bacteriological sampling showed positive results. This did not prevent an outbreak from occurring because of the length of time between sample collection and analysis and subsequent recognition that the positive result represented a real problem. It is often assumed that a positive result is due to accidental contamination in the system. The Interim Standards (7) require check samples to confirm positive bacteriologic results and require State notification within 48 hours. It is unclear whether the 48 hours applies to the results of the check sample or the initial positive sample. Public notification can range from 7 days by radio and TV to the time of the next water bill. If the coliform standard is exceeded, by the time check samples are collected for confirmation, and the maximum time is utilized for notification, it will be too late to take action to prevent an outbreak from occurring. If an outbreak does occur, the only consolation will be that sample results are available to assist the epidemiologist in his investigation. However, with chlorine residual monitoring, some action could be taken immediately by the operator after discovery of a low or zero residual. It is unlikely the operator would assume that he had just made a sampling error when he measures chlorine residual.

Surface water should never be used without treatment, but there are smaller communities and semi-public systems where this is still done. Bacteriological surveillance of these distribution systems can only provide a false sense of security. There can never be assurance that contamination will not occur at any given time even though the bacteriologic results have been negative. Systems using surface water

TABLE 10. STEELE ALABAMA OUTBREAK. 1973

Overall Attack Rate 50/100 (50%)
Infectious Hepatitis

Bacteriological Qualtiy of Distribution Systems

Date	Samples	Results
2/72	1	2/100
2/72-3/73	30	—

Raw water samples 2/72–2/73 15 samples negative for coliforms; 7200 coliforms/100 ml found in late 3/73 during outbreak investigation

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and providing conventional chlorination as the only treatment are not uncommon in some parts of the country. Bacteriological surveillance in these systems is also meaningless, for conventional chlorination can provide a water free of coliforms and yet not be effective in destroying *Giardia* cysts if they are present.

In many instances, ground water free of contamination is used without disinfection. Proper construction and frequent sanitary surveys are required to prevent contamination of these sources. Two examples of outbreaks caused by use of undisinfected ground water illustrate this point (Table 9). The outbreak in the New York State restaurant occurred on November 13–14, 1971, but tap water samples revealed no contamination until November 19. Although source water samples prior to the outbreak revealed coliform contamination, the only treatment provided was softening. A sanitary survey prior to the outbreak would have noted the contaminated source and lack of proper treatment. The Big Sky, Montana outbreak (Table 9) illustrates intermittent contamination of ground water felt to be of good quality and relatively low numbers of coliforms can be associated with illness. The water supply was epidemiologically implicated and chlorination of the source stopped the outbreak. The previous bacteriological history had not predicted the outbreak which occurred in December, 1974–January 1975, for all samples prior to that time were negative for coliforms. Had a complete sanitary survey been performed perhaps the sewage lines passing near the wells would have been noted as a potential hazard as they were during the outbreak investigation.

The remaining outbreaks to be discussed were all caused by treatment deficiencies, and show that routine bacteriological surveillance is of little use in preventing outbreaks due to this cause (Tables 10-12). Frequent sanitary surveys and increased emphasis on maintaining continuous and adequate disinfection are important in preventing outbreaks such as these. Routine bacteriological surveillance data from the Steele, Alabama infectious hepatitis outbreak of February 21–March, 1973 (Table 10) showed relatively little contamination either in the distribution system or the wells. The one positive distribution sample of 2 coliforms per 100 ml would most likely not have caused any concern, as many of us would dismiss this as a sample collector error. Contamination of the limestone aquifer and inadequate disinfection caused the outbreak. Routine bacteriological surveillance was of little help in preventing the outbreak. Monitoring of chlorine residuals, however, might have alerted operators to adjust the level of chlorination during the period of contamination.

TABLE 11. OUTBREAK IN A BOY'S CAMP.
1975

Acute Gastrointestinal Illness – 3 separate outbreaks
Overall attack rates of 12/73 (16%); 85/160 (53%); 67/100 (67%)

Bacteriological Quality of Distribution System

Date	Samples	Results
5/27/75	2	
6/10/75	3	-
6/24/75	3	
7/15/75	3	-
7/29/75	2	
8/26/75	3	
	2	1/100

Raw Water quality 5/75-8/75: 2 samples negative for coliforms

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The outbreak at the Boys Camp (Table 11) is an excellent example of how meaningless the results of a small number of bacteriological samples are in terms of routine surveillance. The bacteriological record for 1975 would certainly not indicate a problem with this supply, yet 3 outbreaks were documented during this period of time in which the water supply was epidemiologically implicated. Samples were collected every month at approximately 2 week intervals, and the outbreaks occurred on June 6, July 18, August 24, all between periods of sampling. The 3 wells used as a water source were chlorinated, but it was reported that the operator did not monitor for chlorine residuals. He relied instead on bacteriological results. Additional information on this system was obtained in 1976 when another outbreak occurred. A thorough engineering evaluation showed one of the wells to be contaminated with 6 to TNTC coliforms per 100 ml and inadequate control of chlorination. The contamination that a routine bacteriological surveillance program failed to uncover was finally found. The outbreaks most likely would have been prevented with a thorough sanitary survey noting geological conditions of the well, etc. and monitoring of chlorine residuals to assist in proper maintenance of disinfection.

The last example, an outbreak at Richmond Heights, Florida, (Table 12) is one with which I am personally familiar. Again routine bacteriological sampling of the distribution system showed no coliform contamination prior to the outbreak. On January 14–15, 1974, chlorination was interrupted and untreated well water was distributed. Routine surveillance noted 3 coliforms per 100 ml in a sample from the system on February 12. The samples collected on February 18 appear to be check samples, but this is not certain. One sample on this date showed 10 coliforms per 100 ml. The 19 negative samples on February 21 appear to be check samples also. During the period of time that check samples were being collected, the community was experiencing an outbreak which affected 1,200 people. The outbreak investigation in March revealed that one of two wells was continuously contaminated by effluent from a septic tank near the well. A sanitary survey and sampling of the raw water could have detected these deficiencies prior to the outbreak and a greater emphasis could have been placed on continuous chlorination and monitoring of chlorine residuals.

Table 12. Richmond Heights Florida outbreak.
1974

Acute Gastrointestinal Illness Compatible With Shigellosis
(10 Culture Proven Cases)
Overall Attack Rate 1200/6500 (18%)

Bacteriological Quality of Distribution System

Date	Samples	Results
1/9/74	9	—
1/10/74	2	—
2/12/74	1	—
2/12/74	1	3/100 ml coliform
2/18/74	1	10/100 ml coliform
2/18/74	2	—
2/21/74	19	—
3/11/74	9	—
3/26/74	2	—
3/28/74	6	—
3/29/74	3	—
	1	>50/100 ml coliform* 3/100 ml fecal*
3/31/74	6	—

SPC - 3/26-31/74 - Average 150/ml
Raw Water - 3/27-4/74 - Total coliform >50/100 ml
Fecal coliform 3 to >50/100 ml

* House with Water Softner

Conclusions

Waterborne outbreaks continue to occur and we should make use of these data to plan and develop surveillance programs. Water supply deficiencies that allowed the outbreaks to occur can be identified and the formulation of control measures should be based on outbreak related problems. Information on etiologic agents can be used to determine treatment requirements. Historical water quality data from systems having an outbreak can be used to determine the effectiveness of bacteriological surveillance.

The data indicate that increased emphasis be placed on surveillance of smaller water systems, especially those serving the travelling public. The formulation of control programs should include increased emphasis on (a) providing adequate treatment of surface water sources to prevent outbreaks of giardiasis, (b) assessing the potential for contamination of ground water sources and providing appropriate treatment, (c) maintaining continuous disinfection, and (d) preventing contamination from entering the distribution system.

Thorough clinical, epidemiologic and laboratory investigations must be conducted to identify possible new etiologic agents responsible for outbreaks of acute gastrointestinal illnesses of unknown etiology. Recognition of outbreaks and timely investigation are necessary for collection and analysis of appropriate water samples and clinical specimens in this regard.

Routine bacteriological surveillance of water distribution systems for coliforms, such as proposed by the Interim Standards (7), is satisfactory for historical purposes but is of little value in preventing the occurrence of waterborne outbreaks. Results of such sampling cannot be received in time for corrective action to be taken. The length of time required for analysis and the time delay between collection and receiving results is much too long to enable the coliform test to be used for routine quality control purposes. The idea of check samples also adds to the time delay, since the first positive sample is generally assumed to be a sampling error rather than real contamination. It is also not logical to permit a lower frequency of sampling for small systems. The ideal quality control program should offer continuous or frequent monitoring that provides an indication of quality within a relatively short time after sampling. In addition to these points, there are instances where negative bacteriological results are misleading and meaningless.

I propose, instead, an alternative monitoring program that would allow our limited microbiological resources to concentrate on research activities, such as identification of etiologic agents responsible for outbreaks of gastrointestinal illness of unknown etiology. The emphasis of this alternative program would be on engineering evaluations or sanitary surveys. A thorough engineering evaluation would include bacteriological monitoring of source water rather than water in the distribution system. It is necessary to know the quality of source water and potential sources of contamination so that appropriate treatment can be recommended. The emphasis then must be on providing and maintaining this treatment, especially continuous disinfection at adequate concentrations. For product quality control, a simple test such as chlorine residual can be used. Of course, chlorine residual must be defined for each system so that all factors of effective disinfection are included, such as time, pH, temperature, species of chlorine, turbidity, and representative sampling points and frequency of collection must be determined. Bacteriological surveillance of the distribution system should be used to check the validity of chlorine residual monitoring, but this can be done at a lower frequency. This would also provide a historical record which many seem to feel is important. If the primary disinfection of choice ceases to be chlorine, residuals or markers of alternative disinfectants would be employed, and the development of these disinfectants should include technology to measure residuals. If supplies are not disinfected, chlorine residual measurements would, of course, not work; however, it is anticipated that a minimum number of such supplies would exist.

The real importance of measuring chlorine residual is that the primary responsibility for taking corrective action is placed where it should be—with the operator. Time is of the essence in preventing an outbreak from occurring and the operator must be the first to know if something is amiss, not the last to know. He can provide the quickest and simplest response simply by adjusting the chlorinator.

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Alternative Indicators of Water Contamination and Some Physiological Characteristics of Heterotrophic Bacteria in Water

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Introduction

The purpose of this presentation is to list some of the most promising microorganisms and procedures that have been suggested to indicate water contamination of public health significance. In addition, studies dealing with the comparative die-off kinetics of selected indicator bacteria and pathogens will be discussed, followed by a brief consideration of reported bacterial growth in certain aquatic environments. These subjects will not be treated exhaustively with a complete listing of literature citations at the end of this paper.

In discussing the question of alternative indicator systems, it is important to list certain criteria that are appropriate in judging their efficacy. Some may seem idealistic, but they are presented within the same altruistic frame of reference that is basic to the indicator concept; that is safe drinking water. The criteria follow:

A. An indicator should be applicable to all types of water that may be investigated.
B. If a microorganism, it should be present in greater numbers than the pathogen in all cases when the latter is found. Alternatively, positive indication should be present if there is a significant health hazard.
C. Any indicator microorganism should not increase significantly in the absence of a health hazard.
D. Indicator microorganisms should be more resistant to the physiological stress within aquatic environments and to the action of disinfectants that are commonly used therein. As a result, the indicators should exhibit greater survival than pathogens under the "real world" condition of the water that is to be tested.
E. The indicator reaction or test data should be unique and characteristic of that microorganism or determination.
F. The indicator methodology should be of minimal complexity, rapid and inexpensive.
G. Indicator mircoorganisms should be harmless to man under usual conditions.
H. The indicator or test should be proportional to the health hazard that is present.

Indicator Microorganisms That Have Been Suggested

In this discussion we have arranged the more commonly used indicator microorganisms first, with others to follows.

A. Coliforms, fecal and *E. coli.*
Since these organisms have already been discussed at this symposium, we have chosen not to emphasize them in this presentation except to mention that some of these microorganisms satisfy most of our criteria

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under a wide range of circumstances. They have also served us rather well for many years (10). However, on occasion they have not been detected when pathogens have been present, as in the case of the Riverside, California, outbreak of 1965, and some authors (4) have attached their efficacy, in part, on the basis of their occasional non-fecal origin in nature.

B. Fecal streptococci.

This group bacteria is frequently used as indicators of sanitary significance However, it must be acknowledged that some species are capable of reproduction exclusive of warm-blooded animals. This was documented in two papers by Mundt in 1963 (15, 16). However, others (7, 18) suggest that fecal streptococci provide valuable supplemental data in determining fecal water contamination if conducted with the fecal coliform test. Such a multibacterial detection scheme could embody the fecal coliform/fecal stretptococcus ratio (8).

C. Standard plate count.

Although it is virtually impossible to enumerate the total population of heterotrophic bacteria within a water sample by a single test, the standard plate count may provide useful water quality data, particularly if it accompanies other complimentary bacterial measurements. The rationale for this lies in the observation that a great many potential or secondary pathogenic invaders that are common to water may be detected in this way. As a result, the overall water quality may be more accurately assessed in addition to the detection of pathogenic bacteria that are known to reproduce in water. Geldreich (9) has supported this contention by adding that a standard plate count limit may also reduce interference in counting low numbers of coliform bacteria.

D. *Clostridium perfringens*

The use of this bacterium as an indicator organism has been advocated by some European workers; most notably, Gunnar Bonde (2). Its spore-forming charactertistic has made it desirable as an indicator in the presence of disinfectants or when water sample transportation time is protracted. In addition, its presence has been correlated with high numbers of classical indicators (2). However, since many authors regard this organism ubiquitous in nature, and it is a strict anaerobe, its widespread usefulness as a sanitary indicator can be questioned.

E. *Klebsiella pneumoniae.*

This bacterium has been employed as an indicator of sanitary significance in Britain and water isolates were recently demonstrated as indistinguishable from clinical isolates of this opportunistic pathogen (20). It is not considered by most to be ubiquitous in nature since it is usually associated with *E. coli,* although it is found in carbohydrate rich wastes and on vegetables and seeds. In addition, it was found in the chlorinated drinking water of Chicago as a fecal coliform (19). It has been pointed out that this organism is capable of proliferating in waters under some environmental conditions (14). This widely held view casts some doubt that *Klebsiella* species may independently serve as definitive indicators although it may be frequently enumerated as a total or fecal coliform.

F. *Aeromonas* species.

Several species of *Aeromonas* are pathogenic to a variety of aquatic fauna and man. These microorganisms are widely distributed in nature, they are considered ubiquitous by some authors (6) and are capable of prolonged survival in many aquatic environments. Because of these qualities, we regard *Aeromonas* species of limited value as indicator organisms in potable water.

G. *Pseudomonas* species.
These opportunistic pathogens represent particular problems in stored water and some medical procedures using water. They are ubiquitous in nature and may actively reproduce in potable and distilled water to populations as high as 10^7/ml (3). Therefore, these microorganisms are a significant health hazard, especially to the hospitalized patient. They may also persist in such water for extended periods of time. Because of this capability, these bacteria are frequently found in the absence of coliforms; one study reported that one-half of the water samples containing *Pseudomonas aeruginosa* lacked coliform bacteria (17).

H. Pathogenic microorganisms.
Some pathogenic microorganisms like *Salmonella* species occur in contaminated water in a statistically reproducible manner if the water is grossly contaminated. However, under conditions where the contamination is present in a lower level, such as potable water, the occurrence of pathogens is less predictable. No single pathogen sufficiently satisfies the criteria of an effective indicator organism. Therefore, until one such pathogen is identified and appropriate detection methods are developed, we feel that the pathogenic microorganisms have many shortcomings as indicators. At some time in the future, developments may allow a pathogen like one of the enteric viruses to serve as such an organism.

I. Others.
Five additional microorganisms will be mentioned. However, the present level of information on each regarding distribution and population dynamics in water is inadequate to make a judgment as to their suitability as indicators. They are *Caryophanon latum*, the *Bifidobacterium*, bacteroides, coliphage and certain acid-fast bacteria.

Other Indicator Procedures: Rapid Tests

The promise of rapid alternatives or supplementary indicator systems is a highly desirous yet elusive goal of water microbiology. This realization awaits the resolution of many theoretical and practical problems through research and testing of procedures such as the ATP biometer, liberation of $^{14}CO_2$ from radio-labeled lactose and the *Limulus* lysate test.

The *Limulus* lysate test for bacterial endotoxin warrants further mention. There is a growing body of evidence that this test could be the most promising of the rapid indicator procedures. Studies that were done by Tom Evans in our laboratories (5) illustrate the relation of this test to some conventional bacterial water quality criteria. The results seen in Figure 1 are typical of our findings and suggest that the *Limulus* lysate assay for endotoxin parallels results of some conventional indicator bacterial tests in natural waters ranging in quality from pristine streams to the discharge of a sewage treatment plant. Answers to questions such as the longevity of endotoxin under conditions of water renovation and disinfection along with further refinement of the assay will bring this procedure within the scope of practical usefulness in the near future.

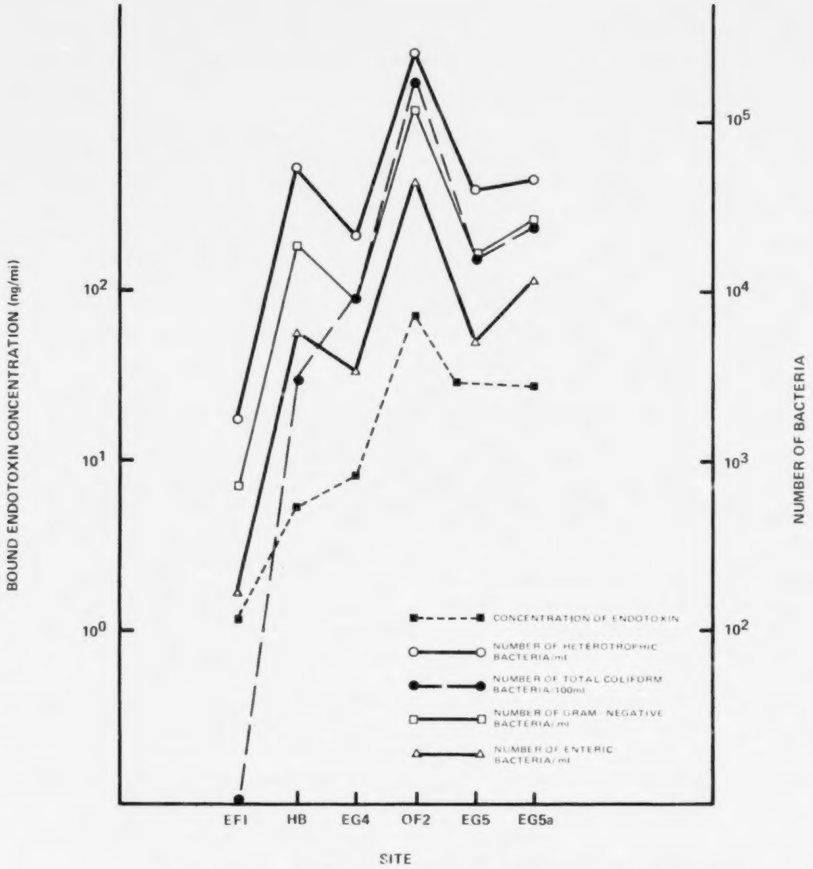

Figure 1. BACTERIAL AND ENDOTOXIN PROFILE OF THE EAST GALLATIN RIVER DRAINAGE FOR SAMPLING DATES 7/11/75 AND 7/15/75. EACH POINT REPRESENTS THE GEOMETRIC MEAN OF FIVE REPLICATE SAMPLES ENUMERATED BY THE SPREAD PLATE TECHNIQUE.

Factors Influencing the Recovery and Enumeration of Indicator Bacteria

An understanding of the population dynamics and physiological injury of heterotrophic bacteria in water is basic to the realistic application of the bacterial indicator concept. This assertion is based on the observation that the aqueous environment, from which the indicator bacteria are recovered, is, at most, an extremely dilute solution of bacterial nutrients. As a result, when bacteria are released into the water from their nutrient-rich natural habitat, they go through a spectrum of debilitation during which enumeration becomes progressively more difficult until they finally die. On the other hand, there is evidence that some heterotrophic indicator bacteria metabolize and even grow in some aquatic environments that are oligotrophic by all other standards.

An overview of these considerations will be given within the context of studies that have been carried out in our laboratories.

In studying the physiological behavior and the population dynamics of heterotrophic microorganisms in water, methodology can be very important. For example, coliform bacteria are capable of cryptic growth, using the organic chemicals liberated from dead cells that have undergone autolysis, if the population is on the order of 10^6/ml in an enclosed container. For that reason we developed the membrane diffusion chamber (Figure 2) to allow the retention of a population of microorganisms within an enclosure that is immersed in a body of water (Figure 3) (12). Water is free to diffuse through the enclosure, carrying remnants of dead cells away and allowing a more realistic experimental approximation of what is likely to occur in the natural aquatic environment. In addition, we found that the population of cells must not be excessive (i.e. less than 10^5/ml) in the performance of these experiments.

Studies were carried out using these chambers to investigate the comparative survival of classical indicator bacteria and some waterborne pathogens in water (13). It was demonstrated that coliforms and enterococci persisted

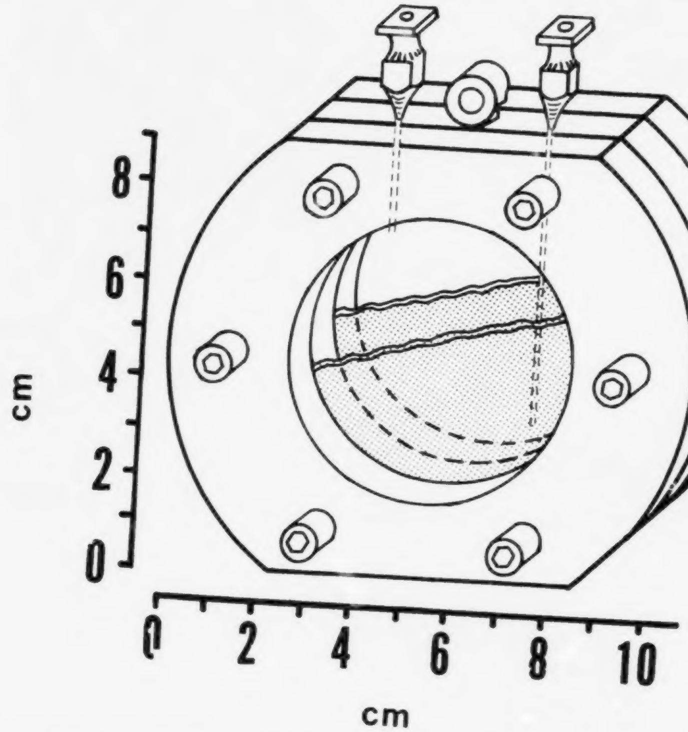

Figure 2. SCHEMATIC DRAWING OF THE MEMBRANE DIFFUSION CHAMBER.

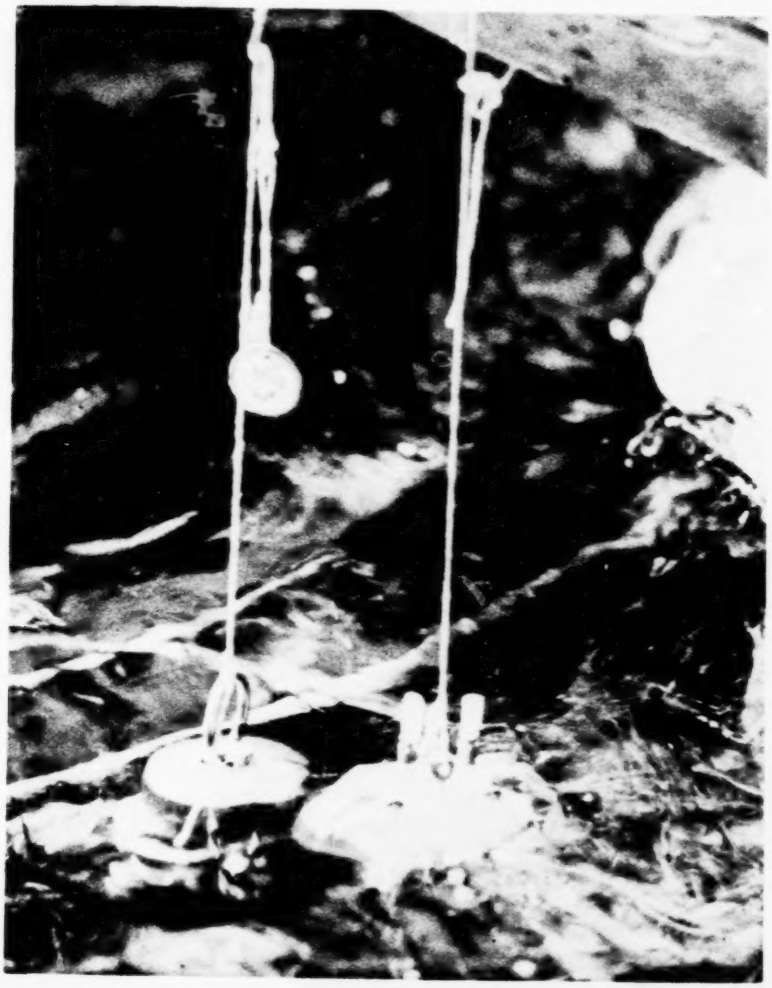

Figure 3. PHOTOGRAPH OF MEMBRANE DIFFUSION CHAMBERS IN OPERATION SUBMERGED IN WATER.

as well as or better than the pathogens that were studied (Table 1). Also, the fecal coliform to fecal streptococcus ratio of human fecal wastes decreased while that from a bovine source increased with time in water to a point where the ratios were not useful in distinguishing the origin of the pollution within 24 to 48 hours. In another study that was carried out in our laboratories, largely by Gary Bissonnette, the physiological debilitation of indicator bacteria in water was examined (1). This work demonstrated that indicator bacteria progressively lose the ability to grow and form colonies on selective media as they age in natural water (Figure 4) and that an appropriate nutrient environment and moderate temperatures allow the repair of such injury within three hours (Figure 5). These studies indicated that this occurrence is widespread among indicator bacteria in nature and that injured cells may comprise as much as 90 percent of the population.

An ongoing investigation describing the interaction between indicator bacteria and algae in the high alpine zone also relates to the broad question of indicator bacterial behavior in water to the collection and interpretation of meaningful bacterial water quality data. Relatively high numbers of total coliform bacteria in a mountain stream have been shown to be associated with an algal mat community. In laboratory studies, indicator bacteria including total coliforms, fecal coliforms and members of the genus *Klebsiella*

TABLE 1. COMPARATIVE DIE-OFF RATES (HALF-TIME)[a] OF FECAL INDICATOR BACTERIA AND ENTERIC PATHOGENS.

Bacteria	Bacteria Half-time (h)	No. of strains analyzed
Indicator bacteria		
Coliform bacteria (avg)	17.0	29
Enterococci (avg)	22.0	20
Coliform from raw sewage	17.5	
Streptococci from raw sewage	19.5	
Streptococcus equinus	10.0	1
S. bovis	4.3	1
Pathogenic bacteria		
Shigella dysenteriae	22.4[b]	1
S. sonnei	24.5[b]	1
S. flexneri	26.8[b]	1
Salmonella enteritidis ser. paratyphi A	16.0[b]	1
S. enteritidis ser. paratyphi D	19.2[b]	1
S. enteritidis ser. typhimurium	16.0[b]	1
S. typhi	6.0	2
Vibrio cholerae	7.2	3
S. enteritidis ser. paratyphi B	2.4	1

[a] The half-time was determined graphically from Fig. 2 and 4 as the time required for a 50% reduction in the initial population.

[b] The half-time was determined graphically from Fig. 4 as the time required for a 50% reduction in the population at 24 h.

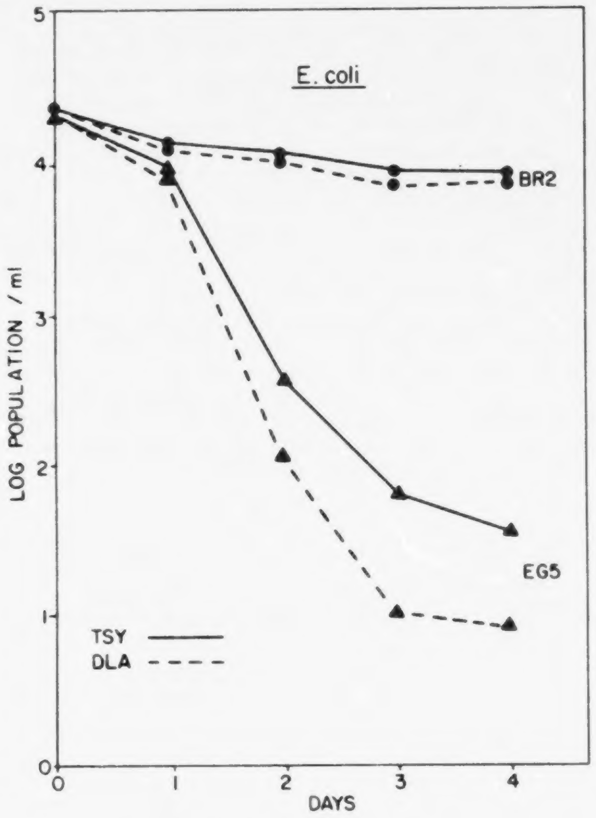

Figure 4. COMPARATIVE RECOVERY FOR E. COLI C320MP25 IN MEMBRANE FILTER CHAMBERS LOCATED AT SITES BR2 AND EG5 DURING A 4 - DAY EXPOSURE PERIOD. SAMPLES WERE SURFACE - OVERLAY PLATED USING TSY AND KF AGAR.

increased by two to three orders of magnitude at low temperatures using the products of algae from the mat community. The likelihood that these bacteria were reproducing this way in nature was further reinforced by the finding that they metabolized radioactive algal organic extracellular products in controlled laboratory experiments. These findings indicate that under some circumstances heterotrophic bacteria are capable of growth in a high quality water. This was previously suggested by Hendricks and Morrison (11) who proposed that stream sediments adsorb organic substances that are subsequently used by bacteria. The implications of these studies go far beyond the alpine streams in which they were done. For instance, surfaces within water storage, treatment and distribution facilities may adsorb and concentrate potential

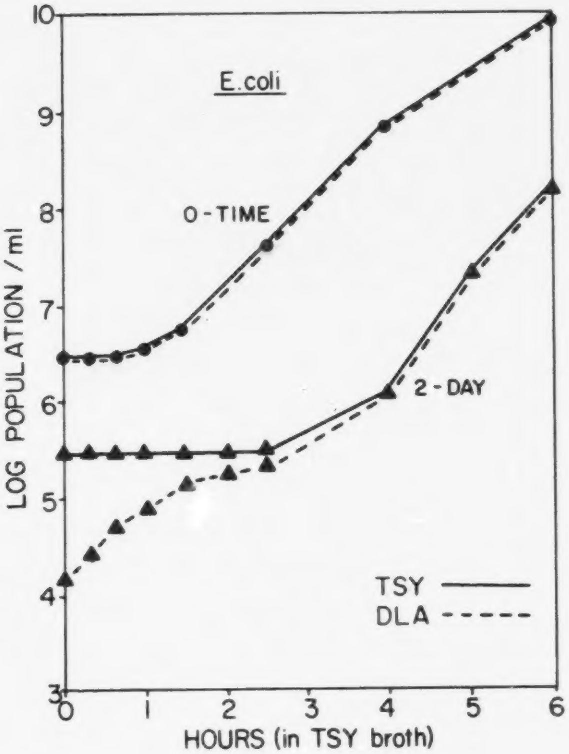

Figure 5. REPAIR OF INJURY IN TSY BROTH FOR E. COLI C320MP25 CELLS HAVING BEEN EXPOSED TO THE STREAM ENVIRONMENT OF SITE EG6 FOR 2 DAYS. CONTROL OR O - TIME CELLS AND 2 - DAY EXPOSED CELLS WERE ENUMERATED OVER A 6 - HOUR GROWTH PERIOD IN TSY BROTH USING TSY AND DLA AGAR SURFACE - OVERLAY PLATES.

bacterial nutrients and provide favorable conditions for bacterial growth both before and after disinfection. Activated carbon filters are an example of an increasingly popular water treatment process that is particularly susceptible to this occurrence. With that in mind, the proliferation of bacterial indicators or potential pathogens in potable waters must be considered in examining alternatives for determining water quality. The growth of indicator bacteria might give a false signal of water contamination, but multiplication of organisms such as *Flavobacterium*, *Pseudomonas*, and *Klebsiella* species, creates a dangerous condition for some consumers that must be recognized if the test procedures are to serve our needs. With the presently used bacterial indicator surveillance protocol, this situation probably does exist in many finished waters that are defined as potable.

Philosophical Basis of Indicator Bacteria and Recommendations

A discussion of the philosophical basis of the indicator bacterial concept is essential in discussing alternative indicator protocols for determining water quality. The philosophical basis that we consider fundamental to the indicator concept is that an indicator system that is more encompassing is better than one that is more restrictive, in terms of the source of the contamination that will be detected. Additionally, false negatives are to be avoided more than false positives if one has to select the lesser of the two evils. As a justification for this position, we cite the distinction between sewage contamination and feces contamination and make the generalization that an effective indicator system will detect not only feces but also sewage contamination. The importance of this notion is that water contamination from sewage may vary widely in amounts of fecal pollution and, more importantly, that the broad spectrum of health threatening chemicals and infectious agents that may be transmitted through water are not exclusively associated with fecal material.

Translated into choices that have been presented, *E. coli* is most restrictive since it is most frequently associated with feces followed by fecal coliforms that are usually linked with fecal pollution, and then total coliforms that may or may not be associated with fecal material. However, total coliforms are a good indicator of sewage or the intrusion of surface or contaminated ground water if found in finished drinking water. As another advantage, the total coliforms are more easily and economically detected with less chance of false negatives than fecal coliforms and *E. coli*. Also, the maximum contaminant level (MCL) for total coliform bacteria might be increased somewhat because of the greater sensitivity in detecting a wide spectrum of contaminant sources plus laboratory contamination with this group of organisms. This would help to satisfy some of the objections to the National Interim Drinking Water Standards that have been voiced in many quarters recently.

Using such an indicator system or even *E. coli*, as in Britain, there is still legitimate cause for concern regarding large numbers of potential pathogens and other microorganisms in drinking water that are capable of reproducing there. This concern was recently expressed by the British (21) in an article that described the proliferation and detection of these organisms within water distribution systems. We propose therefore that a test similar to the present standard plate count be performed as a routine part of the microbiological surveillance of all potable waters and that appropriate MCL's be set. Such a test should be incubated for a minimum of two days at 22 to 30°C to accommodate slower growing bacteria that are adapted to the reduced ambient temperature of their aquatic environment. However valuable the information gained, the time lag required for the performance of this test presents a real logistic problem since each re-test following an excessive result will take at least two days. The implications of this are obvious in terms of the volume of water used in every two-day period. For that reason the utilization of the *Limulus* lysate test would be useful to check for excessive bacterial populations when the standard plate count or coliform counts are high and the operator needs rapid assurance that his remedial action has been effective. This idea may be somewhat futuristic since the *Limulus* lysate test has not yet been perfected or widely accepted. However, we feel that this will become a reality within the next two to five years.

Based on the arguments presented in the preceding sections of this discussion, we propose the following recommendations for a bacterial indicator system in finished water.

A. Total coliforms should be retained as the primary group of indicator organisms with a slight relaxation of that MCL (i.e. less than 3/100 ml).

B. A test similar to the standard plate count should be determined on a regular basis as an index of other microorganisms that are important in water. The MCL level might be near the 20/ml limit that is used in Germany or as high as 500/ml.

C. In the event either the total coliform or standard plate count data exceed the MCL, a *Limulus* lysate test should be performed when it becomes available.

These recommendations represent a realistic yet comprehensive plan to use the enumeration of heterotrophic bacteria and their products to measure the safety of finished drinking waters. We recognize that there are imperfections in this plan, as in others that have been proposed, largely due to the statistical nature of the task and difficulties inherent in searching for heterotrophic bacteria within the hostile and alien environment of treated water. In spite of these difficulties, there is good reason to believe that this protocol will improve the already laudable health safety record of finished drinking water in the United States.

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QUESTION AND ANSWER SESSION

L. J. McCabe, Jr., Health Effects Research Laboratory, U.S. Environmental Protection Agency, Cincinnati, Ohio

What are some of the more significant problems associated with the use of the *Limulus* lysate assay to determine natural water quality over the broad range of ambient waters that might be tested?

G. A. McFeters, Department of Microbiology, Montana State University, Bozeman, Montana

As you have suggested, there are still some potential problems in the application of the *Limulus* lysate test for determining water quality. For instance, we feel that more needs to be known regarding the persistence of endotoxin in waters of various physical and chemical types as well as in water treatment and renovation processes. Possible references of the test due to different water chemistries or additives should also be investigated.

Beyond this, we also acknowledge the need for greater standardization of the assay procedure itself (at present there are four different test kits that are commercially available). Also, more studies need to be performed to allow the correlation of this new test with other established measures of water quality. Such testing should be done under "real world" water conditions.

Some Statistical Considerations in Water Quality Control

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Bethesda, Maryland 20014

As an outsider I am particularly grateful for the opportunity to address you today regarding water quality control. I am a statistician, not a bacteriologist or water quality expert, and have been asked to look at some statistical aspects of water sampling. My analyses are based on the use of the membrane filter method, although they could certainly be applied to other techniques. Indeed, what I have done is an example of the general class of quality control sampling analyses. Each sample must be classified as being either good or bad depending upon whether or not one can detect some relatively rare phenomena. My analyses today are also based upon the idea that the use of coliform indicators is desirable, that they really indicate something we want to know. I do not take into account two facets of the problem that no doubt need ultimately to be considered. They are (a) in measuring a certain number of *E. coli*, using them as indicator bacteria, we have indicated the presence of an uncertain number of pathogens of various sorts and (b) the doses of pathogens needed to induce illness in say 1% of the people drinking water with that level of contamination changes radically from one sort of bacterium to another. Regarding (a), and this is an issue that the experimentalists need to consider, it would be important to have a probabilistic relationship between measured number of *E. coli* and probable numbers of various sorts of pathogens believed to be present.

Specifically, I was asked to investigate three topics. These are (1) how many samples a month should be taken for various size towns, (2) how much water should be taken in each sample and (3) are the present sampling rules well defined? My knowledge of the rules is based upon the EPA interim regulations of December 25, 1976, i.e. specifically, the table appearing in paragraph 141.21 and the instruction of 141.14.

The proposed regulations specify a number of samples that increases steadily with population. It is not evident that this is necessary or desirable. For example, a nationwide voting poll doesn't need substantially more people in its sample than one restricted to a single city. A critical aspect is homogeneity: are we taking a sample from a homogeneous source? If so, many big samples are no better than a smaller number of smaller samples. Figure 1 shows the number of samples per 1,000 population according to the proposed EPA regulations. There are several observations. First, it evidently declines with increasing town size. This seems desirable. There is a small insert in the beginning of the graph; this shows the number of samples per 1,000 population for small towns up to a population of approximately 90,000. The extent of this insert on the larger graph is indicated by a tick mark on the horizontal axis of the larger graph. The insert shows an extremely ragged line, definitely not a consequence of an analytic model. It is not necessarily bad to use seat-of-pants procedures to determine the number of samples per head but in this case it appears not to be rational for small communities.

The next question refers to the amount of water per sample. A standard sample is now 100 milliliters. There is nothing unique about this value other than that it corresponds to a convenient size dish or test tube. Standard statistical theory can tell how large a sample is needed to make sure that we

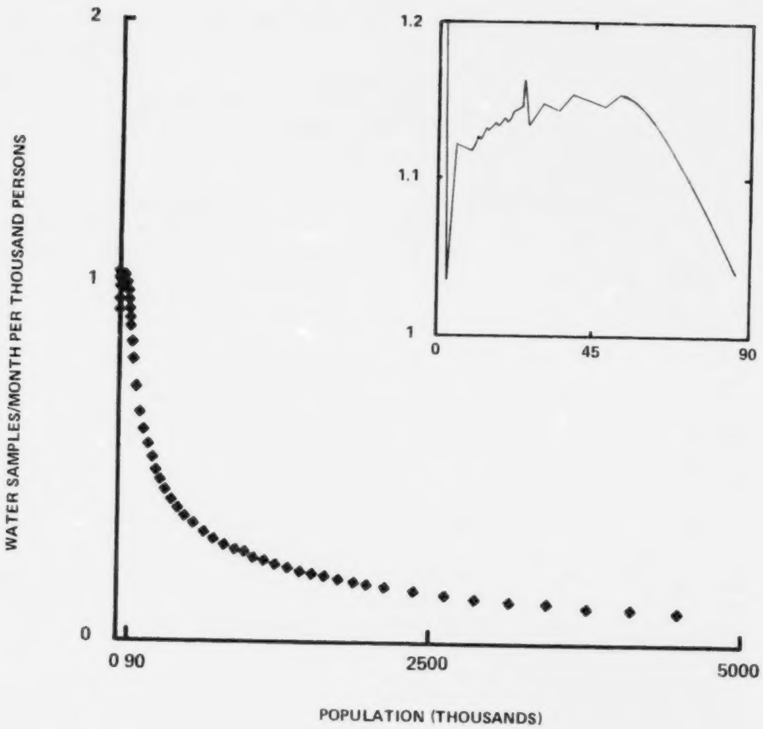

Figure 1. NUMBER OF SAMPLES TAKEN PER 1, 000 PERSONS.

will find something if something is there. Again, the critical issue is the same as previously raised. Do the tubes properly reflect the heterogeneity of the source?

The regulations say that we should examine a certain number of tubes containing 100 milliliters each, that number depending upon community size. For each batch of tubes, say 20 of them, the sampling rule is composed of two parts. First, if the mean number of bacteria per tube is too high, declare the sample to be contaminated. The threshold value is now set at one bacterium per tube. Second, if more than 5% of the tubes contain more than 4 bacteria—even though the overall mean may be less than one—also declare the sample contaminated.

To study this two part sampling procedure we need an analytic model to describe the number of coliform bacteria in a sample. With such a model statistical theory can find the best way of testing the bacterial level of the sample and then the proposed procedure can be compared with the optimal one. It may be that the optimal procedure is excessively complex and the proposed procedure is an effective, simple approximation.

The sampling problem is described by a number of parameters. These are: 1. The level of contamination that we consider just at the border line of acceptable and not acceptable, that is the threshold. 2. A measure of variability of contamination from one water sample to another. 3. The probability that with a given decision procedure and a certain level of contamination that we will "pull the alarm" at exactly the border line level. 4. The probability that we will fail to detect some high level of contamination, one substantially greater than the border line value. 5. The historical information on the frequency of waterborne disease in communities of various sizes.

It is important to distinguish between the case in which the water is uniformly mixed and homogeneous and one in which the water has an important degree of heterogenity. With uniform mixing a relatively small number of samples will suffice to indicate the average number of bacteria per volume of water. In the heterogeneous case we can consider an extreme, indeed unrealistic, example with an extraordinarily high degree of variability. This might be reflected in a situation in which all the contamination was contracted into the form of a bolus. I call this the "bowling ball" model in which unless you hit the particular focal point of contamination with your sampling procedure you will detect no contamination at all. To have a reasonable probability of detection we would need an extraordinarily large number of samples. In the more realistic situation in which contaminants are somewhat clumped it is evident that we will still need a larger number of samples than in the situation of homogeneous mixing.

When the water is uniformly mixed the relevant distribution for the number of particles per standard volume of water is the Poisson distribution which has a single parameter and for which mean and variance are the same. If we assume that this mean varies from sample to sample and can itself be considered a random quantity we arrive at the negative binomial distribution. Professor Pipes of Drexel University and some of his graduate students have considered the practical problem of distinguishing between these two possible distributions, Poisson and negative binomial. Based on a large number of water samples they have almost invariably come to the conclusion that the negative binomial fit the data better than does the Poisson. While this is reassuring it isn't at all surprising since the negative binomial distribution has two parameters and the Poisson only one. Thus we have more flexibility in fitting with the negative binomial. Since the negative binomial arises for the case of the Poisson where the mean is allowed to vary, in the extreme case where the mean doesn't vary we get back to the Poisson, that is, one is a special case of the other. The question then becomes: is the negative binomial distribution enough of an improvement relative to the Poisson to warrant the addition of another parameter? Using the data collected by Dr. Pipes' students I conclude the answer is 'yes'. Making special provisions for a model with a high degree of heterogencity, namely the negative binomial, is justified in real data.

As I have indicated the sampling rule is in two parts, one referring to the mean and one to the number of tubes containing an excessive number of bacteria. As was indicated in a handout received prior to this meeting there are problems with this rule for very small numbers of samples. For example, one bad tube can guarantee that the mean will be too high regardless of zero counts in the others.

Although this rule may not be ideal I have analyzed it using the negative binomial model referred to above. The model is parameterized by a mean level of contamination, a coefficient of variation for the prior distribution of the mean and a sample size, that is, number of tubes. I have calculated the probability that a given water sample will be accepted for various combinations of the parameters. Figure 2 shows the results of a computer simulation.

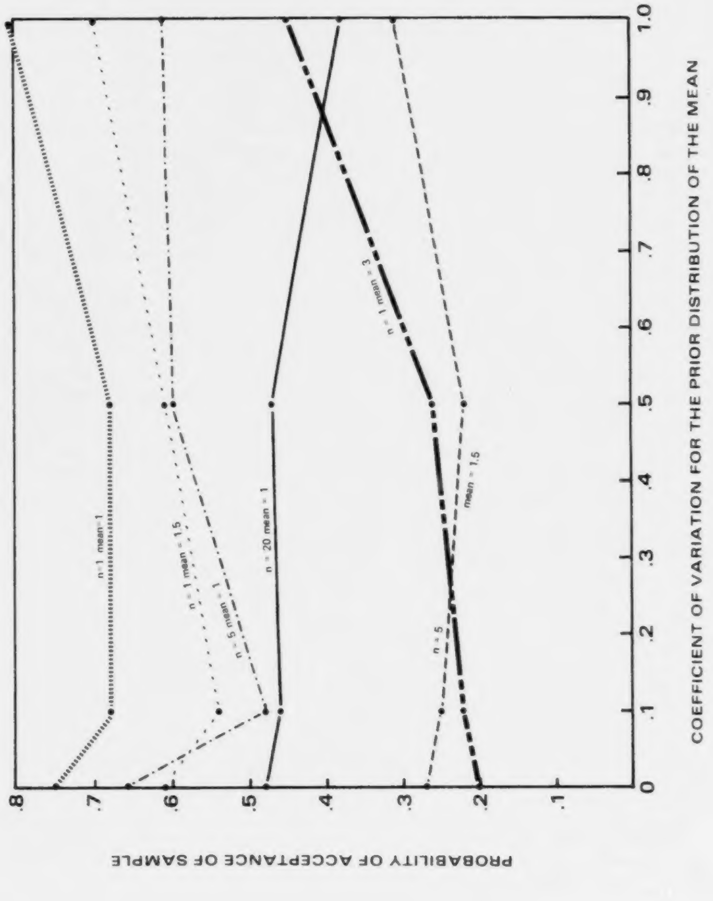

Figure 2. THE PROBABILITY THAT A WATER SAMPLE WILL BE ACCEPTED FOR VARIOUS LEVELS OF MEAN CONTAMINATION, VARIABILITY OF THE MEAN AND NUMBER OF 100 ml ALIQUOTS; USING THE PRESENTLY PROPOSED EPA RULE AND COMPUTER SIMULATION. THE MEAN IS IN UNITS OF BACTERIA/100 ml.

Because I have used a simulation rather than exact calculations the answers have a great deal of variability themselves and sometimes behave counter-intuitively.

Some general observations are that: 1. There is still a high probability of accepting a relatively bad sample, one with a mean of three coliform per 100 mls., if the number of tubes is small enough. 2. There is a general tendency for the acceptance probability to increase as the variability of the mean increases. This is entirely reasonable if we think back to my bowling ball model. 3. Things do behave in a rational fashion in that the probability of acceptance goes down with increased mean and with increased numbers of tubes.

If my model for bacterial distribution is a reasonable representation of reality then this rule is not the optimal one. It is suboptimal in the sense that for a given probability of considering uncontaminated samples to be contaminated we could achieve a higher probability of correctly detecting a contaminated sample with some other rule. Unfortunately the optimal rule is a good deal more complicated. It says "reject when the mean is too high, but the threshold that defines too high must be based on the variance of the number of bacteria found per tube."

The last topic I want to consider is historical data regarding the level of water-borne disease by community size. The goal of quality control is to

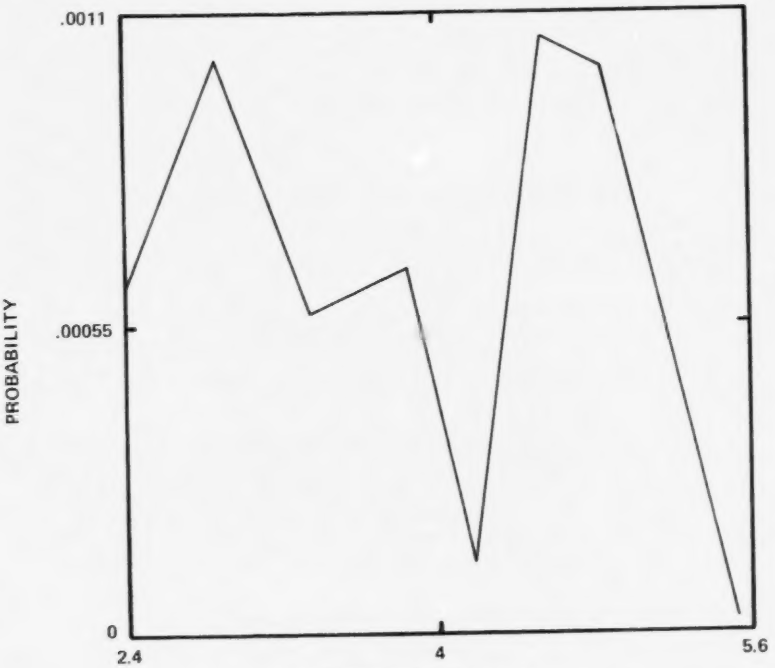

Figure 3. PROBABILITY ANY ONE PERSON WAS INVOLVED IN AN OUTBREAK OF WATERBORNE DISEASE DURING 1946 - 1970.

keep to a low level the probability that some person will contract water-borne disease. In a community with a poor historical record for the incidence of such disease we would have to be more stringent with the sampling rules in order to keep the probability down to the same level for all citizens in all communities.

The historical record is a confusing one. My last figure shows the probability that a single individual will be involved in an outbreak of water-borne disease at some time in the time interval 1946-1970 as a function of community size (on a $\log_{10}$ scale). Small towns (3-5 thousand) and rather large towns (50-100 thousand) have higher rates than towns of other sizes. A possible explanation for this is that the very smallest towns don't need elaborate quality control mechanisms so they have a low disease rate. Larger towns (the first peak in the curve) do need such surveillance but don't introduce it until town size is near ten-thousand. Towns of 50-100 thousand population need quality control of a higher order of sophistication but this isn't to be found (the second peak) until town size approaches one-half million. Of course, this reasoning is entirely a product of having seen the data first.

If we accept the odd behavior of this curve as representing reality we see that there could be objections to a sampling plan which weighted the intensity of sampling using historical information. From a political viewpoint, it might be difficult to justify less sampling for towns of 10-thousand population than for those which are either smaller or larger.

After all this, please note that the maximum value of the line indicates a probability of less than one part in a thousand over a 25 year span.

I urge you to remember that sampling and quality control are problems to which the statistician can make a major contribution and his advice should be solicited regarding any proposed regulations.

QUESTION AND ANSWER SESSION

S. M. Morrison, Department of Microbiology, Colorado State University, Fort Collins, Colorado

How does your model take into account heterogenity with respect to small and large towns that have their own treatment and distribution systems?

L. Muenz, National Cancer Institute, National Institutes of Health, Bethesda, Maryland

It doesn't do so in any explicit way. The coefficient-of-variation parameter can be adjusted to agree with historical information from a town of a given size but this sort of adjustment is not built into the model; it's at the discretion of the user. A more complicated model would already have the community size-variability relation explicitly included although I wonder how useful such a mixture of mathematics and survey information would be.

P. D. Haney, Black and Veatch, Kansas City, Missouri

In the 1940's the U.S. Public Health Service published a graph giving the relation between number of water samples/month and population. J. K. Hoskins, who was Assistant Surgeon General at the time said that the mathematical derivation was so complicated that they only published the curve and not the mathematics. Perhaps you could track this down.

L. Muenz

My plot shows the number of samples/month/1000 persons as a function of community size using information from the E.P.A. interim regulations. Assuming that this is the same number of samples/month as referred to in the 1940's there is no way to ascribe a mathematical origin to this curve. The demonstration of this is given by looking at the "ink blot" of points corresponding to very low populations. This region is blown up in the insert and a tick mark (′) on the horizontal axis of the larger graph shows how far the insert extends. The behavior of the insert is completely non-analytic even if most of the larger graph seems rather smooth. No conceivable mathematical function could give rise to the jagged line appearing in the insert. Thus the whole curve is likely to be empirical; this, of course, doesn't mean that it's bad, just that it would be difficult to defend.

Chlorine Residual Substitution — Rationale

Leland J. McCabe
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Consideration of the use of chlorine residual as an indication of the safety of a drinking water supply was much easier before the discovery of the chloroform problem. In the past a chlorine residual was an unquestioned good. Now the value of the residual must be determined so that trade-off considerations can be given to the possible increase in chloroform and the other trihalomethanes. Both toxicological and epidemiological studies indicate an adverse effect of chloroform ingestion or the consumption of water that has been chlorinated.

The value of carrying a disinfection residual to overcome contamination throughout the distribution system of a community water supply has not been determined. Johns Hopkins University now has a research project to determine the effectiveness of the residual to contain introduced contamination to a distribution system (3). The value of the residual in a special situation has been reviewed. It is believed that residual chlorine is a substantial safety factor on shipboard distribution systems but credible evidence is not available to insist that such a residual be maintained (2).

Besides the safety factor that a disinfection residual may provide to a distribution system there is another benefit that must be considered. If the water supply operations is set up to maintain a residual, the absence of the residual can quickly signal the inadequacy of treatment or the introduction of foreign material into the distribution system. Thus, the measurement of a disinfection residual can provide a continuous-real-time monitoring of the safety of the distributed drinking water.

To control the spread of waterborne disease, information on water quality must be available in time to influence decisions concerning the use of the water. Microbiological methods are not adequate as they only provide historical data on the quality of water several days ago. Microbiological methods can be very helpful in the investigation of the causes of waterborne disease outbreaks but they do not lend themselves to monitoring.

The need to rethink what we have been calling water quality monitoring or surveillance was demonstrated in the Community Water Supply Survey (4). The survey design called for sampling all water supplies in one metropolitan area of each of the Public Health Services regions. A late change was made in Region I to include the State of Vermont instead of New Haven, Connecticut. Some virus studies were being conducted on the New Haven supply and it was thought best not to include this supply as part of the national survey. Nationally, it was found that at 85 percent of the water systems, bacteriological samples were not collected at the rate prescribed in the drinking water standards. Table 1 is a copy of the table from the report of the survey. Samples were collected at less than half of the prescribed frequency at 69% of the supplies surveyed. The prescribed frequency is admittedly an arbitrary number but when efforts were made to examine the required frequency the established frequency was universally supported by the state sanitary engineers in 1968. The results of the bacteriological sampling are shown in Table 2 and it can be seen that the problem of poor

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TABLE 1. BACTERIOLOGICAL SURVEILLANCE EVALUATION BY COMMUNITY SIZE.

	Population Served in Thousands (All data are per cent of System Totals)							Overall System Totals	
	≤.5	.5-5	5-10	10-25	25-50	50-100	>100	Number	Per Cent
Met bacteriological surveillance criteria	*4*	*18*	*12*	*10*	*3*	*12*	*36*	100	*10*
Did not meet bacteriological surveillance criteria	*96*	*82*	*88*	*90*	*97*	*88*	*64*	869	*90*
Did not collect samples at the rate in the bacteriological surveillance criteria	*94*	*74*	*83*	*85*	*92*	*88*	*64*	827	*85*
Systems totals—number	446	315	75	59	36	16	22	969	—

Note: 60 per cent of the survey population were served drinking water by systems over which a bacteriological surveillance program met the criteria.

TABLE 2. WATER SUPPLY SYSTEMS IN VARIOUS POPULATION SIZES WHERE DELIVERED WATER EXCEEDED THE COLIFORM DENSITY LIMIT.

Population Range	Number of Systems	Number of Systems Exceeding	Per Cent of Systems Exceeding	Per Cent of Survey Population Exceeding
≤500	446	79	*18*	*15*
501–5,000	315	29	*10*	*8*
5,000–10,000	75	5	*7*	*7*
10,001–25,000	59	5	*9*	*7*
25,001–50,000	36	1	*3*	*3*
50,001–100,000	16	1	*6*	*6*
>100,000	22	0	*0*	*0*
Totals	969	120	*12*	*2*

quality water is greater with the smaller supplies. Something needs to be developed that can address that problem.

The usefulness of the chlorine residual to maintain the water within the bacteriological limits of the standards was also demonstrated in the CWSS. Although the practice of chlorination caused a dramatic decline in the frequency of the samples containing coliforms, these organisms are not eliminated merely by claiming that chlorination is practiced. Unless a chlorine residual was maintained in the distribution system, a significant portion of the samples from the distribution system contained coliforms. The coliforms are eliminated nearly completely in systems providing at least a trace of chlorine residual (1).

Buelow and Walton (1) also analyzed Cincinnati's water surveillance data, during a period when a free chlorine residual was being maintained in the plant effluent. This is illustrated in Figure 1 when the free residual was carried in 1969-1970. All monthly average coliform counts were below 0.1 per 100 ml. and in most cases the samples with coliforms were collected from locations without chlorine residuals.

If the reality of these data is faced, it is obvious that something new must be done in the way of improving bacteriological surveillance. The idea of substituting the chlorine residual for coliform testing emerged from considering the results of the Community Water Supply Survey. The concept was outlined by Robeck (5) but he cautioned that five specific problems should be addressed before the substitution is allowed for a water supply.

1. The number and location of samples for which chlorine residuals are to be substituted.
2. The form and concentration of chlorine residuals to be maintained.
3. The frequency of chlorine residual determination.
4. The analytical method to be used.
5. The past bacteriological history of sampling location.

The problems were considered and paragraph(h) of Section 141.21 of the National Interim Primary Drinking Water Regulation (6) allows for the substitution of chlorine residual for 75% of the microbiological sampling. At least 4 chlorine residuals are substituted for each microbiological sample dropped but the residual must be measured daily. The benefit of the use of this provision to the safety of the smaller water supplies is obvious. Daily monitoring of a water supply would not be achieved by the coliform test,

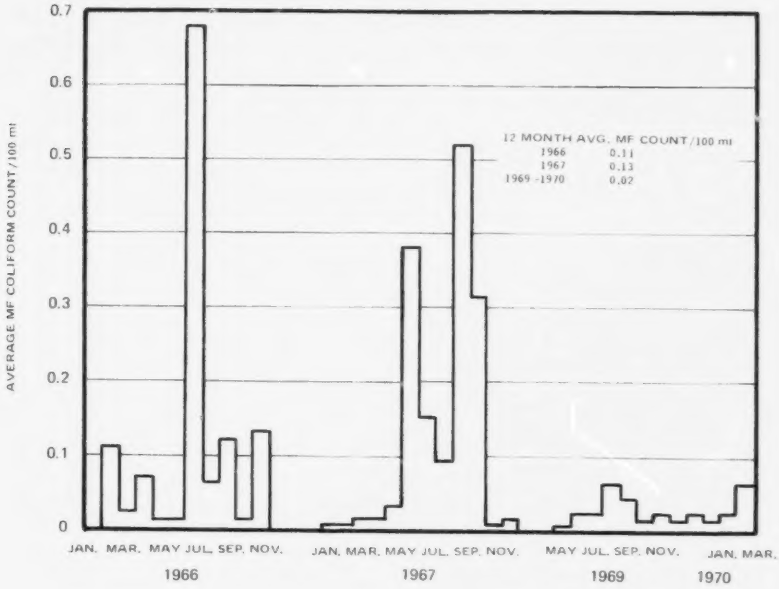

Figure 1. MONTHLY AVERAGE COLIFORM COUNTS PER 100m1.

unless the supply served 25,000 people. For a population served of 25,000 to 28,000, 30 microbiological samples are collected per month.

If the rules are followed the chlorine residual procedure will assure much quicker action in correcting a problem of unsafe water. "When a particular sampling point has been shown to have a free chlorine residual less than 0.2 mg/1, the water at that location shall be retested as soon as practicable and in any event within one hour." Thus, the operator has one hour to increase or restore the chlorine application and make sure the residual reaches the sampling point. With coliform testing, considering sample transit times, time to perform the test, and time to make the notification, the reaction to bad water might be half a week's delay. A good example of the response following the loss of chlorine residual was the investigation of a cross connection in Tampa, Florida (Communication from Hillsborough County Health Department to the State Division of Health, November 27, 1973). A pressure sewer had been mistakenly connected to the water main and the location of the problem was accomplished with the chlorine residual testing.

Chlorine residual testing could have been required in addition to the required microbiological monitoring but to promote the use of the more effective monitoring it was decided to eliminate the more costly microbiological testing. Savings effected can be used for other water quality testing. Responsibility for first line quality control must rest with the water works operator. The present system of collecting limited samples at small water supplies and sending them to the state laboratory for testing introduces a mystical aspect to quality control. The operator is not involved personally and reports come back a week or more later that may indicate deficiencies in the past. When the operator determines a chlorine residual and does not find a pink color

(DPD test), he is the only one to know there is a problem and he must make a decision to protect the public's health. The responsibility for an acute problem is where it should be and at the only place where necessary corrective action can be taken. The publication of the causes of waterborne outbreaks must be intensified to reinforce the responsibility of the water-works operator in the prevention of disease. This will require the reporting and investigation of outbreaks and this activity can be supported with EPA's water supply program grants. The state laboratory microbiologist should no longer be overburdened with routine bacteriology samples and should be able to participate in outbreak investigations to determine etiological agents and their source. Table 3 shows a tabulation of waterborne disease outbreaks by agent. The large percent attributed to the nonspecific category gastroenteritis points out the need for more extensive microbiological workups during outbreak investigation.

Section 141.21 paragraph (h) requires the collection of coliform samples when the chlorine residual cannot be restored within one hour. This sample has a great likelihood to contain coliforms but it is to be averaged in to indicate the monthly means for compliance with section 141.14. This provides a much stricter level of quality for a supply using the chlorine residual procedure. For example a water supply serving 5,000 population is required to collect 6 microbiological samples and may substitute the chlorine test for 4 of them. Thus 2 coliform samples would be collected per month but surveillance would be provided daily by the chlorine residual test. If the residual were low on one day of the month and the follow up coliform count was 4 per 100 ml the system would not meet the drinking water standards even though the supply likely had good quality water all but one day of the month. To promote the use of the chlorine residual substitution, which provides really a better monitoring system, the days with adequate chlorine residual should be considered zero coliform days in computing an average.

TABLE 3. WATERBORNE DISEASE OUTBREAKS DISTRIBUTION BY ETIOLOGY.
1946-1974

	Community Water Systems %	Other Water Systems %
Gastroenteritis	52.6	47.5
Infectious Hepatitis	16.3	13.7
Shigellosis	9.6	10.2
Chemical Poisoning	5.9	4.0
Giardiasis	5.2	2.5
Typhoid	4.4	15.8
Salmonellosis	4.4	2.8
Amebiasis	0.7	1.2
Poliomyelitis	0.7	0.0
Enteropathogenic *E. coli*	0.0	1.2
Tularemia	0.0	0.6
Leptospirosis	0.0	0.3
Number of Outbreaks	135	322

Recommendation

I would think that complete chlorination substitution would be usable for the smaller supplies. If this were allowed for supplies up to 9400 population served it would also take care of the averaging problem with a small number of microbiological samples. A bacteriological sample is required when the chlorine cannot be restored within an hour. The operator of the system would of course continue to work to get the residual restored and it will have to be assumed that this will be done as diligently as possible. When he gets the residual back up to the prescribed level the public is again protected. What would be the best microbiological test to perform on this sample that was taken before the problem was corrected? A sample for *Clostridium perfringes* would seem to provide data on what was needed to be known and the sample could be just mailed to the laboratory without a concern for transit time. These organisms are more resistant to chlorine and should be ubiquitous enough to indicate when polluted water enters the distribution system.

For large water systems the full benefits of the chlorine residual should be promoted. Continuous monitoring is possible with feedback controls to adjust the treatment and give alarms when departures occur. In these systems the microbiology applied to samples collected when the residual chlorine cannot be restored within one hour should be rather complete. The testing should assist in determining the source of contamination and ought to be more elaborate than just a coliform test.

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QUESTION AND ANSWER SESSION

R. R. Colwell, Department of Microbiology, University of Maryland, College Park, Maryland

How can chlorine residuals supply information concerning actual bacteria populations in drinking water? If chlorine is that effective in the treatment of pathogens in sewage, why do we need to chlorinate the drinking water if we do it to sewage?

L. J. McCabe, Health Effects Research Laboratory, U.S. Environmental Protection Agency, Cincinnati, Ohio

The chlorine residual test is not supposed to give a quantitative evaluation of the bacterial population. When the chlorine residual exceeds the specified amount it is an immediate qualitative evaluation that pathogens are not present.

Pathogens are also derived from non-point sources that are not treated. It will be a great day when all sewage is disinfected adequately and with reliability, until this Utopia, it would seem desirable to disinfect drinking water. A residual disinfectant provides some measure of protection in distribution systems.

W. Ginsberg, Water Purification Laboratories, Chicago Bureau of Water, Chicago, Illinois

Do you believe water treatment plant operators are likely to take four times the number of chlorine residual samples when they often do not take a sufficient number of coliform samples now?

L. J. McCabe

We want to provide to the operator the most effective test for him to use. Their use of any test is a matter of personal modification and enforcement. I think these would be enhanced with a chlorine residual measurement. In the case where neither test were used, the chlorine residual requirement would waste less money because it would not be necessary to have an unused microbiology capability standing by. With the use of the chlorine residual, the microbiological capability can be used for more in depth analyses of the most meaningful samples and be ready for waterborne disease outbreak investigations.

Public Health Considerations of the Microbiology of "Potable" Water

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Water is essential for every biological function and the convenience of obtaining a ready supply of drinking water from municipal supplies is accepted without question in civilized societies. In fact, the occasional odor of chlorine when a tap is turned on has been interpreted by the layman as a guarantee that the water has been purified and is, therefore, safe to consume. Unfortunately, there are problems, other than microorganisms, as, for example, taints, odors, floccules and disturbing colorations, which cause temporary distress and, perhaps, a telephone call of complaint to the supplying agency; otherwise, in North America water is available and used without further thought.

Natural waters which flow in rivers and streams, and settle in wells and lakes, contain a distinctive flora and fauna, and characteristic dissolved and undissolved inorganic and organic matter. Some components of the microflora are potential pathogens and, therefore, hazardous to the health of the community whereas other microorganisms are beneficial in mineralizing organic matter, concentrating heavy metals in trickling filters, and acting antagonistically to many of the potentially pathogenic bacteria. It is the function of water supply agencies to provide "clean" water in quantities necessary to cope with the demand of the community. This water is collected from rivers and wells and sometimes from estuaries or the sea. The water that is collected is treated with flocculants, e.g. alum, to precipitate suspended material, filtered through sand beds to remove precipitates and flocs of living organisms, and chlorinated to eliminate potentially harmful organisms. Additional compounds often are added to the water (such as fluorides) and the finished water is distributed to storage reservoirs or through pipes to consumers. The water is not microbiologically sterile since spores and many Gram-negative rods survive chlorination, and it is not pure, as fluorides and other compounds may be added. However, the quality of the finished water should be safe for drinking and usage in hospitals, pharmacies and industry. Periodically, samples of water are collected from the distribution system and chemical analyses done, but, unfortunately, rather primitive techniques are used to assay microbiological quality. The microbiological analyses are recognized to be time-consuming and may provide questionable information, particularly since the few bacteriological tests that are most commonly done are performed by inexperienced or inadequately trained personnel. Very rarely is a registered microbiologist employed to carry out the bacteriological tests.

Increasingly, problems with drinking water are being brought up for public discussion. Setting aside the problems of toxic chemicals and speaking only to microbiological criteria for water quality, there are several problems that need to be addressed, as, for example, the difficulties in establishment of standard laboratory procedures, standardization of laboratories, and development of criteria for qualification of professional environmental and

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sanitary microbiologists. The major problems are, clearly, (1) standardization of personnel and training; (2) standardization of laboratories carrying out the analyses; (3) standardization of reporting results and banking of the data for coordination and intercalibration around the country; and (4) standardization of media and reagents, as well as procedures for sample gathering, plating, culturing, reading tests, interpretation of test results, and recording of data. Clearly, the need for professionalization of water testing and water quality determination is most urgent. Papers in this series deal with this problem and it can be seen that there are solutions, but that effort, time and money will be required.

The microbiological standard for drinking water has not changed in its essentials since 1914 (10), and is determined by reference to the presence of coliform bacteria. The definition of coliform is vague and includes many asporogenous, Gram-negative rods (21). Their isolation from water is dependent upon the antiquated Most-Probable-Numbers Technique (1) or by the newer mebrane-filtration method (1). The presence of "coliforms" in numbers greater than 1/100 ml (27) is taken to mean that the water is contaminated. The solution is often re-chlorination of reservoirs and pipe water, and the problem, thereafter, quickly forgotten. It takes at least 48 h for coliforms to be recognized by the standard procedures employed (23), by which time the contaminated water will have been distributed and consumed by many individuals in the community. Thus, at best, the bacteriological methods presently employed provide only an indication that trouble occurred in the system and the event may have been 2 or 3 days ago, hardly to be considered satisfactory water quality control. Thankfully, water-borne infections are rare in the U.S.A., even though the potential exists for dissemination of disease-producing microorganisms. Thus, modern microbiology techniques should be incorporated in routine examination of water. Gas-liquid chromatography appears to offer immediate detection of bacteria and bacterial products by direct analysis. Methods such as pyrolysis and laser scanning also offer potential value in direct assessment of water quality. The application of molecular genetic techniques to water bacteriology appears to be almost totally lacking. Reasoner (23) correctly stated that "the development of quantitative procedures for pathogenic bacteria has lagged behind the development of quantitative procedures for non-pathogenic bacteria." Unfortunately, it can also be stated that water bacteriology has not advanced to a degree commensurate with that of other aspects of microbiology, most notably of microbial genetics and physiology. If the aim of water quality determination is to locate the presence of pathogens or indicator bacteria, then it is essential not only to be aware of other types of organisms occurring in water, but also to understand their physiology, ecology, and role in public health.

Bacterial Microflora in Treated Water

Although it is relatively easy to remove some microorganisms from water by filtration and chlorination, it is difficult to reduce the content of organic material to a level below which it will not support microbial growth (2). Attempts to sterilize water supplies by means of disinfectants are generally ineffective and invalidate the description of pure water (13). The viable, aerobic, heterotrophic bacteria which can be routinely isolated from treated water include *Achromobacter, Arthrobacter, Bacillus, Clostridium,* coryneforms, *Flavobacterium, Gallionella, Klebsiella, Leptothrix, Micrococcus, Mycobacterium, Proteus. Pseudomonas* and *Serratia* (4, 26, 29, 30, 32). These survive inadequate treatment or enter via contamination within the distribution system. Although none of these organisms are primary pathogens, some may be responsible for taints and odors, spoilage of commercial products for which water is used, and also may act as secondary invaders in human disease processes.

Pseudomonas and *Flavobacterium* are problematic genera, some species of which are opportunistic pathogens (14). The latter genus, a heterogeneous taxon of convenience (3, 20), comprises a variety of species, some of which are tolerant to high levels of chlorine, with certain strains resisting as much as 100 mg/L. *Pseudomonas* and *Flavobacterium* spp. have been noted to cause heavy growth in the sprinkler heads of scrub sinks and water baths in hospital operating theatres and preparatory areas (15). Yellow-pigmented rods, described as flavobacteria, have been isolated from patients with systemic infections following heart-lung surgery. One species, *F. meningosepticum,* is relatively resistant to antibiotics (22) and its presence should not be tolerated in treated water. The dangers resulting from bacterial metabolites, such as pyrogens, should not be overlooked, and in one study (24) it was demonstrated that water used for pharmaceutical purposes contained viable bacteria producing pyrogens. The problems caused by bacteria in water extends to scientific laboratories where, deionized and soft water units have been shown to contain pigmented Gram-negative rods. In particular, bacterial growth occurs in the resin columns, with the bacteria population comprising as many as 10^5 cells/ml (25). Surely, methods must be devised to ensure the sterility of all water that is to be used as drinking water.

Purification of Water

The effectiveness of chlorination justifiably can be criticized because of the resistance of many viruses and bacteria to chlorine treatment. Treatment with ozone is considered to be an alternative method of water treatment since ozone is bacteriocidal and removes taints and odors without leaving unpleasant residues (11). Methods not presently employed in water treatment, but of potential value, include heat sterilization and membrane filtration, both of which can be adapted for purification of large volumes of water. Reverse osmosis and ultrafiltration are perhaps the most feasible and effective in reducing bacterial numbers (6). In all cases, except autoclaving and ultrafiltration, viable organisms remain in the water after treatment and this fact raises several questions (13):

(a) are the microorganisms remaining in drinking water after conventional treatment merely harmless indicators of an unsterile condition?
(b) are they beneficial by assisting in the breakdown of organic compounds transported by the water?
(c) are they a hazard to the system, the user or as contaminants in an end product of industrial processes intended for ingestion or injection by humans?

Microbiological Problems with Water Treatment and Distribution

McCabe and co-workers (19) found that, of 969 public water supply systems serving 18.2 million individuals, 613 utilized ground-water systems, 67% of which distributed water to 2.8 million consumers without any disinfection. Of 621 wells, 9% produced water that contained coliforms, 2% contained fecal coliforms, and 11% of the wells dispensed water with >

500 heterotrophs/ml as determined by plate counts. How many pathogens, particularly organisms causing gastro-enteritis, were distributed in those waters can only be speculated.

It is particularly disquieting to realize that even water in hospitals is far from pure. In one interesting study (18) it was recorded that as much as 30-40% of water contained in so-called sterile tanks and pipes serving operating theatres were contaminated, due largely to improper maintenance.

Microbiological examination of water, as presently carried out in most municipalities, comprises the Most-Probable-Numbers technique for coliform determination, membrane filtration, and total counts of viable bacteria using plate count methods. The incubation temperature routinely used is *ca* 35-37°C, and little concern is shown for organisms which need lower or higher incubation temperatures. How do we know that *all* coliforms grow at 35°C? In a study in our laboratory, it was found that a greater number of tubes of lactose broth gave positive (acid and gas) reactions when incubated at 20°C than at 35°C. It is conceivable that the lower temperatures permit the multiplication of organisms better suited or adapted to the aquatic environment. However, these may also be coliforms, and should be considered as "environment-adapted" coliforms. Suppression of coliforms by the microbial flora of a given environment can also occur. For the latter, false negative results will be recorded giving an erroneous impression of water purity.

The problems of plasmid transfer, particularly of lac + plasmids, need to be carefully examined. In particular, Orndorff *et al.* (21) recovered many lactose-fermenting, i.e. acid and gas producing, *Pseudomonas* spp. from aquatic samples. The sanitary significance of these organisms is unclear, except that false reactions, interpreted to signify the presence of coliforms, will be obtained when such organisms are present in water being tested for potability or as a source of drinking water. Plate counts, if done properly, give an indication of the total number of bacteria in water; sudden increases in numbers indicate trouble somewhere in the system. Yet, few of the smaller water treatment plants attempt to quantify the total bacterial populations in the water (7).

It is important that the initial treatment of water be adequate to reduce, or preferably eliminate, bacteria and viruses. Good flocculation is necessary for optimum virus reduction (7). The filter beds must be carefully managed to prevent "floc" breakthrough. Development of filters capable of retaining amoeba cysts is essential, since conventional disinfecting procedures are ineffective against removal of these organisms. Furthermore, harmful bacteria can be transported via the cysts.

At the present time, the problem of viruses in drinking water is vexing. It is disputed whether viruses can, indeed, be isolated from drinking water. It is obvious that as methods for concentrating, culturing, isolating, and identifying viruses occurring in natural waters are developed and improved, this aspect of drinking water quality will be better understood and dealt with on a rational, objective basis.

Chlorination is regarded as the saviour of water quality. Yet turbid waters contain particles with embedded bacteria that can be protected from the effects of chlorination (7). Indeed, coliform bacteria have been found to survive in turbid waters chlorinated to provide 0.1-0.5 mg/L of free residual chlorine after contact for 30 min. It should not be overlooked that open reservoirs of treated waters may receive inputs of fecal matter from birds and wild animals. Thus, if water is treated only before it enters the distribution system and not carefully monitored, en route, it may be several days before a sanitary problem is detected, when it may already be too late for action to avoid health hazard.

Tap Water

Bacteriological examinations of tap water frequently reveal the presence of large numbers of bacteria. Coliforms and fecal streptococci occur infrequently in chlorinated water, and from their presence is inferred 'recent' contamination.

In a bacteriological study of tap water undertaken in our laboratory, (Table 1), it was shown that samples collected in the Washington, D.C. area were negative for coliforms, as determined by incubation for up to 7 days at 35°C in lactose broth. However, after this extended incubation time the plate counts on glucose tryptone yeast-extract agar (GTYEA) were alarmingly high, although lower than for bottled water (Table 2). After incubation for only 48 h these plate counts were very low, yet the prolonged incubation period at 25°C was necessary for development of the majority of the bacterial colonies. This trend was repeated, in part, on Eosin-Methylene-Blue agar (EMB), which allows the selective growth of coliforms. Several samples (Table 1) contained β-haemolytic colonies on blood agar incubated aerobically, and anaerobically under hydrogen. Thus, it is recommended that plate counts be done at 20-25°C, the incubation periods be increased to 7-14 days, and a wider range of bacteriological media be employed in routine examination of water.

Drinking Fountains

The convenience of drinking fountains for providing refreshing, cool water is accepted as part of everyday life, yet the bacteriological standard of the water at public drinking fountains is low. The drinking fountain head, unless regularly cleaned, is often dirty. In 1965, 78% of drinking fountains tested (12) were contaminated, yet in 1975, 76% were in a similar poor state. It is interesting to note that a wide range of microorganisms were recovered from the fountains. Although it is assumed that drinking water always contains a residual chlorine level, the water from the drinking fountains tested by Herman (12) was found to be devoid of the disinfectant. This may be explained by the separate water delivery systems for drinking fountains, in which the water is cooled and constantly circulated, with the result that the chlorine level usually falls below the effective level of 0.6 mg/L, thus permitting a diverse range of microbial growth at the air-water interface on the fountain head. Clearly, the bacteriological quality of water at communal drinking fountains needs to be constantly monitored and, where necessary, the water delivery systems modified.

Bottled Water

Bottled water has gained popularity in the U.S.A. with annual sales exceeding $150 million (31). In 1972, there were an estimated 700 water bottling plants (8), with California the largest customer. The quality standards of bottled waters varies between states, with some complying with the Public Health Service Water Standards (22), whereas others have lesser or no standards. However, from the bottle labels it is often implied that the quality is superior to the municipal water supply, and that this quality will

TABLE 1. BACTERIOLOGICAL EXAMINATION OF TAP WATER OBTAINED FROM THE DISTRIBUTION SYSTEM WITHIN THE WASHINGTON, D. C. METROPOLITAN AREA[a]

Source	Presumptive coliforms/ 100 ml	No. of colonies/ml							
		GTYEA[b]		EMB[c]		Hektoen Enteric Agar	Sheep Blood Agar		
							(Aerobic)		(Anaerobic)
	after 7 days	2 days	7 days	2 days	7 days	7 days	2 days	7 days	3 days
U.S. Capitol	0	5	7.8×10^2	0	60	0	5	10	40
U.S. Supreme Court	0	20	1.4×10^4	40	4×10^3	0	30	30	70
Washington-Suburban Sanitary Station	0	0	1.1×10^2	0	5	0	0	5	0
Alexandria City Hall	0	0	7×10^3	0	9×10^2	0	0	0	0
Fairfax County Offices	0	5	4×10^2	0	1.2×10^3	0	0	0	0
Montgomery County Offices	0	10	50	0	0	0	20	30	60
Prince Georges Gounty Court House	0	0	0	0	0	0	0	0	0

[a] Water was allowed to run for 2-3 min before collecting samples in sterile bottles. Inoculated media, except for the presumptive coliform test, were incubated at 25°C for the time periods indicated.

[b] Glucose tryptone yeast-extract agar (Difco).

[c] Eosin methylene-blue agar (Difco).

TABLE 2. BACTERIOLOGICAL EXAMINATION OF BOTTLED WATER. INCUBATION OF MEDIA, EXCEPT THE PRESUMPTIVE COLIFORM TESTS, WAS AT 25°C.

		No. of colonies/ml								
	Presumptive coliforms/ 100 ml	GTYEA [a]		EMB [b]		Hektoen Enteric Agar		Sheep Blood Agar		
								(Aerobic)		(Anaerobic)
Source (Brand name)	after 7 days	2 days	7 days	2 days	7 days	2 days [f]	7 days	2 days	7 days	3 days
Aqua Pure [c]	0	10^3	10^2	0	0	0	0	50	50	0
Deer Park	0	4.4×10^6	con [d]	2×10^5	con	5.1×10^2	6.4×10^6	2×10^6	con	1.1×10^6
Great Bear	0	5×10^2	1.5×10^6	6.3×10^5	6.6×10^5	0	0	3×10^3	5.4×10^4	3×10^3
Mountain Laurel Spring Water2 [e]	0	0	50	2×10^3	2×10^2	0	0	1.5×10^3	1.5×10^2	6×10^3
Mountain Valley Mineral Water	0	0	0	0	0	0	0	0	0	0
Poland Water obtained from:										
Grocery Store	0	4×10^6	con	5×10^4	7×10^4	0	0	1.7×10^4	1.8×10^4	0
Manufacturer	0	6.6×10^5	con	3.5×10^4	3.5×10^4	0	0	1.5×10^4	1.5×10^4	0
U.S. Supreme Court	0	1.8×10^6	con	1.8×10^4	1.8×10^4	0	6×10^2	3.7×10^4	6.4×10^4	1.8×10^3

[a] Glucose tryptone yeast-extract agar (Difco)
[b] Eosin methylene-blue (Difco)
[c] Pasteurized
[d] con = confluent growth
[e] From hot springs, >90°C
[f] Refrigerated

be maintained throughout the shelf life of the product. Unfortunately, this is usually far from true, and the bacteriological quality of freshly bottled water varies from brand to brand (9). Furthermore, the bacterial count tends to increase with storage. The usefulness of the bottled water standards adopted by the Food and Drug Administration, which became effective on May 22, 1974 (28), is questionable. They do not include a plate count limit for bacteria, but the coliform limit of less than 2.2 organisms/10^2 ml is similar to the drinking water standard. It is a help that bottled water is categorized as a food, but it is a pity that bottlers often recommend it for preparing baby food, pharmaceutical prescriptions and hot and cold beverages. Thus, certain members of the community, namely the young, aged and infirm, may be more susceptible to infection by opportunist organisms which could occur in the water.

In a study undertaken in this laboratory, the bacteriological quality of bottled water obtained in Washington, D.C. was examined (Table 2). Although the coliform numbers, as determined by the production of acid and gas (in lactose broth) within 48 h, were negligible; in some cases there were large numbers of aerobic heterotrophs as determined from plate counts made on GTYEA medium. The significant point to be made is that, except in the case of the coliform tests, the test media were incubated at 25°C. When incubated at 37°C, little or no growth was obtained. Thus, it is misleading to enumerate bacteria in water by incubating the inoculated test media at 37°C.

It can be seen (Table 2) that pasteurized water, i.e., Aqua Pure, and refrigerated mineral water was of good bacteriological quality; however, the numbers of bacteria present in other brands was disturbingly high. These data were obtained several years ago (*Washington Star*, Washington, D.C., July 11, 1971). Monitoring of the microbiological quality of bottled water should be done on a regular basis to ensure the public health safety of this product. Of particular concern was the large number of β-haemolytic colonies which developed on blood agar. These colonies may reflect the presence of potentially harmful organisms in the water, and the numbers greatly exceed those found in tap water (Table 1). Furthermore, it is readily obvious that the bacterial counts for stored, bottled water, viz., Poland water obtained from a grocery store and from the U.S. Supreme Court at the time the study was carried out, were higher than in water obtained directly from the bottler. Clearly, long-term storage of bottled water can have an adverse effect in providing for growth of bacteria. It would seem appropriate for bottlers to either pasteurize the water, or require refrigeration during storage.

Recommendations

Too little is known about the microbial ecology of water at source, treatment, distribution and storage. It is essential that the normal microbial flora of drinking water be regularly monitored, and the taxonomic composition of the microorganisms present in source water be determined on a regular basis. Thus, seasonal variations in the flora can be measured so that alterations in the flora can be recognized to be abnormal increases in the total number of microorganisms or selective increases in a few taxa due to seasonality.

It is desirable not to have any viable organisms in drinking water, and methods to improve the treatment of water, including removal of viruses, and maintain its purity until consumption, should be employed in water treatment systems.

Methods presently employed for the examination of water are inappropriate and give a poor idea of water quality. It is important to examine the effectiveness of a wide range of media, such as blood agar and low nutrient media, pre-treatment at low temperatures, and of using a wide range of incubation temperatures, especially low temperature incubation of 20-25°C for enumeration of total viable bacterial populations. Thus, psychrophiles, mesophiles and thermophiles should be considered because of the variety of conditions and uses for drinking water. Non-selective enrichment techniques have been successful in the recovery of *Salmonella* spp. (17) and *Vibrio cholerae* (5) from the Chesapeake Bay, and these methods may be effective in determining the sanitary quality of drinking water. It is important to remember that no single technique is suitable for the isolation of all pathogens; therefore, a combination of methods is recommended, with the minimum being inclusion of total viable counts at 25°C.

The concept of a residual chlorine level in drinking water needs to be carefully scrutinized because of chlorine resistance amongst viruses and many bacteria. Although chlorine-resistant coliforms may not occur, the presence of *Flavobacterium* and *Pseudomonas* is undesirable and represents a potential health hazard. Examination of drinking water for the presence of *Pseudomonas* should be incorporated in the routine analyses of the water since a suitable method exists for the enumeration of *P. aeruginosa* (16). Also, other *Pseudomonas* spp. are reasonably easily identified and enumerated.

The taxonomy of *Flavobacterium* must be radically improved, and isolation procedures for pathogenic species of this genus should be developed.

The problems appertaining to antibiotic resistance and plasmid transfer between taxa should be studied and the significance of these phenomena to public health safety determined.

In summary, the present methods for bacteriological analysis of drinking water, although unsatisfactory in many respects, should *not* be discarded in favor of the chlorine residual or other such chemical test. Rather, the bacteriological testing should be improved, expanded and modernized. It is never wise to throw the baby out with the bath water, and substitution of the chlorine residual for sound microbiological analyses would do just that!

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Water Analysis by the Membrane Filter Procedure

SESSION 2

Coliform Detection and Control

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Sampling Collection and Handling

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The ideal transported water sample is one which reaches the laboratory with its bacterial flora unchanged, both qualitatively and quantitatively, as it was at the time of collection. The type and degree of change is best determined by analyzing the samples at the time and location of collection for comparison of results obtained by the same procedural analysis after transit. Unfortunately this action is impractical for a routine drinking water monitoring program. Changes which may occur are dependent upon many variables and combinations of the individual variables. Factors which influence the bacterial flora during transit are quality of the sample bottle, bacterial population, density of each type comprising the population, concentration of nutrients both organic and inorganic, fluctuations in temperature during transit, turbidity, clumping and antagonistic action; all of these exhibit their effect on the bacterial flora during the transit/storage period. These factors can and do vary for every water sample, regardless of water classification as to source, surface, ground or potable water. Sampling locations must be carefully selected to insure representative sampling, collection procedures must be conscientiously followed to prevent accidential contamination, and every attempt must be made to transport samples promptly to the laboratory for processing. Failure to comply with these facts regarding sample collection and handling will produce data which are confusing, misleading and detrimental to any monitoring program.

Collection of Potable Water Samples

Proper collection procedures and techniques are fundamental to obtaining reliable results for any water sample. Errors relating to sampling are usually greater than those associated with laboratory analysis.

The laboratory is usually responsible for supplying sterile sample bottles. Each bottle must be properly prepared with respect to the type water being sampled with thiosulfate added to bottles for sampling chlorinated water supplies, or a chelating agent (EDTA) for waters containing metals, copper and/or zinc. Quality control tests must be conducted on each batch of bottles sterilized to assure sterility of all bottles. Sterile bottles should not be stored more than 30 days to reduce possible contamination due to accidental damage during storage. After sample collectors receive sterile bottles, it is their responsibility to prevent accidental contamination during the collection procedure. Bottles must not be carried in vehicles unprotected and certainly not for several days to even weeks. Samples should be collected from a distribution system tap which does not have any attachments. Taps which have anti-splash devices and/or home purification attachments should be avoided or removed before sample collection. It should be known that the tap is supplying water from a service pipe directly connected with the main and is not served from any type of storage tank (1,2,5). The tap should be opened and water allowed to run to waste for 2 to 3 minutes, or for a time sufficient to clear the service line. The recommended time for flushing varies from 2 to 5 minutes. Flushing time is relative; the intent is to flush the service line to

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assure that the sample collected is representative of water in the main. Treatment of water taps before collecting potable water samples is unnecessary if water is allowed to run to waste and reasonable consideration given to the choice of sampling taps (5). Thomas *et al* (13) reported that flaming taps resulted in no significant differences for both total and fecal coliforms nor for the standard plate count.

The sample bottle must remain closed until it is to be filled. The stopper must be protected from contamination when removed and during the actual sample collection. The bottle should be filled without rinsing, leaving an ample air space so that the sample can be thoroughly mixed before analysis, and the stopper replaced immediately (1,5).

Compliance with recognized sample collection protocol can prevent accidental contamination and reduce the occurrence of spurious samples. Sample collection is an important part of the testing procedure; the test is no better, nor more reliable than the sample which arrives at the laboratory for analysis. Laboratories must be certified, evaluated, approved—choose your word—the intent is the same; however, there are few programs which provide adequate and repeated training for sample collectors. Such training is recognized as worthwhile but economically impractical; whereas failure to have such a program may really be the economical impractability. Serious consideration must be given to the implementation of a program designed to train and periodically recertify sample collectors, similar to the milk program.

In application, transit time and storage are so interrelated, they should be considered together. Standard Methods (1) provides recommendations for transit/storage times for stream, source and potable water samples. Stream and source water samples should be held below 10°C during a maximum transit time of 6 hours. Such samples should be refrigerated when received in the laboratory and processed within two hours. When local conditions dictate delays in sample delivery longer than 6 hours, consideration should be given to field examination or by use of the tentative delayed-incubation total coliform procedure. In a study on sample storage time-temperature effects, Geldreich *et al* (6) observed that MPN coliform counts conducted on samples held at 5°C for periods up to 48 hours were in acceptable agreement with initial MPN results than counts observed on samples stored at 13-32°C or at 35°C. Also, they observed better stability for samples held at 5°C and examined by the MF technique. The more precise results obtained by the direct MF count as compared to the statistical MPN estimate revealed a more subtle change in bacterial densities during storage. These recommendations and information basically apply to stream and source water samples. Since these requirements may not be realistic for individual potable water samples transported to the laboratory by mail service, the time interval between collection and analysis should in no case exceed 48 hours, preferably within 30 hours as specified in Standard Methods. Even the recommendation for refrigeration for potable water samples can be interpreted as optional and not a requirement.

Presently, most State programs do not enforce the 30-hour transit/storage time limit. Because of a multitude of reasons, e.g. inefficient mail service and inability to utilize other means of transportation due to geographical locations of remote water supplies, State programs allow examination of samples which arrive within 48 hours or more realistically, within two calendar days. Few, if any State programs require refrigeration of potable water samples during the transit period.

Conscientious investigations were conducted by many workers in an attempt to determine a transit/storage time of water samples which would not adversely affect the bacterial flora either qualitatively or quantitatively. Cox and Clairborne (4) reported that refrigerated water samples showed relatively little change after 48 hours, while paired samples stored at room temperature showed a noticeable change. However, the lowest coliform density obtained at zero hours storage was 390/100 ml which would not qualify their samples as potable water. Shipe and Fields (12) indicated that a chelating agent (EDTA) added to stream samples known to contain copper and zinc, 0.16 mg/l and 6.5 mg/l, respectively, reduced the rate of decrease in *E. coli* density. They concluded that this chelating agent would be of value in maintaining the coliform index near the level existing at the time of sample collection for a storage period of 24 hours. Data indicated that addition of EDTA at the time of sampling resulted in approximately 69% of the cells remaining viable after 2 hours and 40% after 24 hours. However, there is no indication of survival of coliform organisms in water when the initial density was between 1 and 10/100 ml. Calwell and Parr (3) and Hoather (8) reported a gradual decrease in coliforms with increased storage time whether samples were iced or not. Lucking (10) concluded that the time element between collection and examination is important and testing must begin within 48 hours of collection. Lansome *et al* (9) concluded that samples having a coliform density between 1-20/100 ml need not be refrigerated up to a 48-hour storage period with no adverse effects on coliform detection.

Previous investigations relating to transit/storage time of samples have omitted information regarding the influence of increased non-coliform densities on coliform survival and detection, especially with respect to potable water supplies. However, Lucking (10) and Lansome *et al* (9) certainly recognized this as a factor directly relating to the over-all problems of transit and storage. They also recognized that this and other factors such as changes in temperature, availability of nutrients, oxygen tension and chemical concentration are extremely varied in widely separated geographical areas, therefore making the transit/storage problem unique for each water supply. Excessive non-coliform densities may interfere with coliform detection when examining potable waters expected to have low coliform densities (7). Analyses of bacteriological data from a survey of distribution water transmitted by 969 public water plants did suggest that high non-coliform densities can interfere with detection of low levels of coliform. Data in Table 1 shows that the frequency of detecting total and fecal coliforms increases as the standard plate count increased to levels up to 500/ml; but decreases in frequency when the non-coliform densities exceed 1000/ml. These findings indicate that high densities of non-coliform bacterial populations may adversely affect coliform detection. The factor of high density non-coliform organisms versus low density coliform organisms is just being investigated and may be correlated to transit/storage time for potable water as well as stream and source water supplies. Data obtained on the distribution system water quality in Northern Kentucky reflects the potential interference of coliform detection exhibited by high densities of non-coliforms after a 24-30 hour transit/storage period. The Northern Kentucky treatment plant laboratory data, based on samples examined less than six hours after collection, indicated satisfactory water quality throughout the distribution system. Data obtained on similar samples collected during the same sampling period and from the same distribution system but examined after a 24-30 hours transit time, were reported by the State Laboratory as being potentially unsatisfactory. Results of these samples showed excessive or confluent bacterial growth. In some instances sheen was

TABLE 1. BACTERIAL PLATE COUNT VS. COLIFORM DETECTION IN DISTRIBUTION WATER NETWORKS FOR 969 PUBLIC WATER SUPPLIES.

*General Bacterial Population **		*Total Coliform*		*Fecal Coliform*	
Density Range per 1 ml	Number of Samples	Occurrences	Percent	Occurrences	Percent
<1 - 10	1013	47	4.6	22	2.2
11 - 30	371	28	7.5	12	3.2
31 - 100	396	72	18.2	28	7.1
101 - 300	272	48	17.6	20	7.4
301 - 500	120	30	25.0	11	9.2
501 - 1,000	110	21	19.1	9	8.2
> 1,000	164	31	18.9	5	3.0
TOTAL	2446	277	------	107	------

* Standard Plate Count (48 hrs. incubation, 35°C)

observed associated with the confluent growth, but distinct coliform colonies did not develop which prevented quantitation of the coliform density. Unpublished data obtained during a study of this Northern Kentucky distribution system indicate a statistical significant increase of the non-coliform density after a 24-hour storage period. The density increase appears to be temperature dependent and is greater during the summer months when the distribution system water temperature reaches 26°-27°C. This increase in non-coliform density occurs even though the free chlorine residual is maintained at a concentration usually above 1 mg/1 throughout the distribution system.

Limited data is available relating to transit/storage time specifically for potable waters. However, the data available does indicate that samples examined immediately after collection will provide the most valid results. Since this is impractical for potable water, data indicate that such samples should be examined as soon as possible after collection. The transit/storage period ranges from 6 to as many as 72 hours. However, 24 hours for potable waters appears to be acceptable by the majority of investigators while a few indicate that 48 hours is also acceptable. All of the studies do indicate that the transit/storage problem is dependent upon a multitude of factors and the influence of these factors, individually or combined, will vary for every water examined, be it stream, source or potable.

The bacteriological flora of a water sample will either increase or decrease upon storage. Which change and the degree of change are difficult to predict and will vary with each sample, water supply and geographical location. If these changes, which are related to transit/storage, result in unacceptable data, there are alternate steps which should be considered. Samples could be sent to laboratories nearer the water supply. Additional laboratories or regional laboratories could be established. Alternate means of transporting samples other than the mail service, such as bus lines, private courier or United Parcel Service, should be seriously investigated. The delayed membrane filter total coliform test could also be considered, however, on a case by case basis and not adopted as a cure-all procedure.

Perhaps the transit/storage dilemma for potable water cannot be resolved until research is conducted dealing only with potable water samples. Such a research project must address many types of water supplies, in many geographical areas to obtain results which can resolve the problem. Until definitive data are compiled, it is necessary to make recommendations based upon the data presently available, taking into consideration the conflicting interpretations of that data as applied to potable water samples. A strong recommendation is made that potable water samples be processed within thirty hours after collection; this is a maximum transit/storage time period. If the laboratory is required by State regulations to examine samples after 30 hours and up to a 48-hour transit/storage period, the laboratory should indicate that the data may not be valid because of excessive delay before sample processing. Samples arriving for processing after a 48-hour period should be refused without exception and a new sample requested.

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The EPA Methods Manual and the Microbiological Standards

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Uniform application of the best available methods is essential for generating the valid data needed to enforce water quality standards. The EPA methods manual, *Microbiological Methods for Monitoring the Environment, Part I Water and Wastes* (2) provides these methods in support of the special needs of the Agency.

The purpose of this report is to preview the microbiology manual in preparation, show how it supports the Agency's responsibility to monitor the quality of our Nation's waters, and demonstrate particularly how it performs this function for the National Interim Primary Drinking Water Regulations (NIPDWR).

Responsibility for Standardization

The Federal Water Pollution Control Act Amendments of 1972, the Marine Protection, Research and Sanctuaries Act of 1972 (commonly referred to as the Ocean Disposal Act) and the Safe Drinking Water Act of 1974 direct the Environmental Protection Agency to protect and improve the quality of our water resources and to control pollution. To achieve these goals the Agency must set, enforce and monitor standards for water supplies, ambient waters and wastewater discharges. In EPA, the Environmental Monitoring and Support Laboratory (EMSL)—Cincinnati is responsible for selecting, validating and standardizing analytical methods for water and for establishing quality control procedures.

With the publication of the microbiological methods manual, EMSL—Cincinnati has completed its mission to provide standardized methods for chemistry, biology and microbiology. In addition, EMSL has prepared a quality control handbook for water and wastewater laboratories.

These manuals are not replacements for nor in competition with *Standard Methods for the Examination of Water and Wastewater,* (1) but rather provide the basic methods for EPA to carry out its enforcement responsibilities. The EPA microbiology manual cites as valuable sources of further detail and background information *Standard Methods* and EPA's *Handbook for Evaluating Water Bacteriological Laboratories* (3).

Development of the Manual

The standardization process for microbiological procedures began at a seminar held in San Francisco January, 1973 where the Agency's needs for standard microbiological methods were agreed on and a steering committee was formed to develop the EPA manual. It represented the Office of Research and Development, the Regional Offices, the Office of Water Programs and the National Environmental Investigations Center (Office of Enforcement). Committee members were assigned responsibility for preparing sections on selected microbiological parameters. First drafts were presented and reviewed at the second meeting of the committee in January, 1974 at Cincinnati where the basic design, format and content were also established for the manual.

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The editors, Robert Bordner and John Winter, EMSL—Cincinnati, developed, formatted and edited these materials and added technical detail and information reflecting Agency policy. This draft was reviewed by EPA programs and Regional offices. The many constructive review comments, changes and corrections are now being incorporated into the final version of the manual which is scheduled for distribution in September, 1977.

Legal Authority

The manual will be the basic reference for monitoring water and wastes in compliance with applicable water and effluent standards established by the Agency. Publication in the Federal Register will establish the legal authority for the methods used in the NPDWR program as was done for the procedures used in the National Pollution Discharge Elimination System (NPDES) for the analysis of effluents.

The Code of Federal Regulations (CFR) Title 40, Part 136 (6) lists the procedures required for Section 304 (g) of the Federal Water Pollution Control Act (NPDES). Table 1, an excerpt from that Federal Register, shows the approved test procedures for bacterial parameters. References are made to *Standard Methods for the Examination of Water and Wastewater* (1) and the methods of the U.S. Geological Survey (5). When released, the EPA microbiology manual will be referenced in the column provided in the table. The amendments also provide blanket approval of alternate test procedures by applications to EMSL—Cincinnati and for individual approval by application through the EPA Regions.

It is anticipated that the alternate test procedures for the Naional Primary Drinking Water Regulaions will be referenced similarly by amendment to the Federal Register.

Objectives of the Manual

To carry out the legal requirements set forth in the three water quality acts the EPA manual must provide:

1. *The best available standardized methods for water quality determinations, enforcement actions, compliance monitoring, resource planning and research activities.* The procedures will be the reference methods for the evaluation of modified or new methods.

The accomplishment of the objectives of the manual will assure the uniform application of the most reliable procedures in all laboratories. Uniform procedures facilitate comparison, interchange and common use of data in cooperative field surveys, interlaboratory studies and information storage and retrieval systems.

2. *Methods that have the general consensus of microbiologists within EPA.* Many methods are in use by researchers or by water analysts that have not been proven by application in the various waters and geographical areas of this country. These methods are not necessarily acceptable for use in the EPA.

3. *Methods that are tailor-made to meet the specific needs of the Agency.* Procedures acceptable for some agencies may not meet the needs of EPA. For example, if a decision is made for more stringent holding time requirements for potable water samples than has previously been practiced, such specification will be put in the manual.

TABLE 1. LIST OF APPROVED TEST PROCEDURES FOR WASTEWATER *

Parameter and units	Method	References (page nos.)			
		EPA methods	14th ed. standard methods	PT. 31 1975 ASTM	USGS methods [7]
BACTERIA					
4. Coliform (fecal)[5], number per 100 ml.	MPN; membrane filter		922		
5. Coliform (fecal)[5], in presence of chlorine, number per 100 ml.	do [6, 8]		937	45	
			922		
			928,937		
6. Coliform (total)[5], number per 100 ml.	do [6]		916		
7. Coliform (total)[5], in presence of chlorine, number per 100 ml.	MPN; [6] membrane filter with enrichment.		928	35	
			916		
			933		
8. Fecal streptococci[5], number per 100 ml.	MPN; [6] membrane filter; plate count.		943		
			944	50	
			947		

[5] The method used must be specified.

[6] The 5 tube MPN is used.

[7] Slack, K.V. and others, "Methods for Collection and Analysis of Aquatic Biological and Microbiological Samples: U.S. Geological Survey Techniques of Water-Resources Inv. book 5, ch. A4 (1973)".

[8] Since the membrane filter technique usually yields low and variable recovery from chlorinated wastewaters, the MPN method will be required to resolve any controversies.

* FEDERAL REGISTER, VOL. 40, NO. 232-40 CFR 136, DECEMBER 1, 1976

4. *Procedures with limited options.* For enforcement purposes the analyst is required to use one preferred procedure whenever possible. At the First Microbiology Seminar on Standardization of Methods in San Francisco (4) in 1973 Jeter reported four variations of membrane filter tests and sixteen ways to conduct multiple tube tests for total coliforms in *Standard Methods.* The EPA manual must provide a single, direct method wherever possible. For example the use of lactose broth for the MPN analysis of total coliforms will not be included in the EPA manual.

5. *Validation of methods previous to publication in the manual.* Criteria for validation are specified for new or additional methods to be included in the manual. No methods will be added to the manual without sufficient supporting data.

6. *Quick response to changing needs of the Agency is possible by the single agency action of EPA.* For example the Water Supply Laboratory Certification Program requires criteria and procedures to evaluate all water supply laboratories. This very timely and nationally-important information was inserted in the microbiology manual without delay. If the Maximum Contaminant Levels or methods of establishing levels for total coliforms in drinking water should change, the manual could easily reflect these changes.

Special Features of the Manual

The EPA manual features several special characteristics:

1. A looseleaf style is planned for the manual so that method improvements or additions can be made easily. For example, a recent development in the manufacture of membrane filters produced a filter of larger pore size for microbiological analyses. A statement was added to the manual allowing the use of this filter, which is not permitted by *Standard Methods.* If the National Academy of Science recommends additional parameters for drinking water, for example the standard plate count, the methods would be added to the manual.

2. New editions are planned as demanded to keep the contents of the manual current. These revisions can be published in as little as 18 months. As improved test procedures are developed and validated and other parameters are proved useful they will be considered for the manual.

3. The manual guides the analyst in selection and use of the best method for his needs. In response to the recent controversy concerning use of the membrane filter procedures for chlorinated effluents, the EPA manual limits the methodology.

4. Important supporting information for good laboratory practice is included in the manual. Examples are a section on quality control of microbiological procedures, an area that has been largely ignored. Another section is devoted to laboratory and field safety. A third applies basic statistics to microbiological analyses, and a fourth discusses legal considerations for the microbiologist.

5. The manual will be widely distributed to Federal, State, local and private laboratories performing water analyses.

6. It is written in a simple, direct, stepwise style that is easily understandable and prepared in a "cook book" form so that it can be used at the bench by the analyst. The looseleaf form will also be conducive to use at the bench.

7. The format assigns a decimal number to each paragraph for easy reference and location, similar to the American Society for Testing and Materials Books of Standards.

Summary

In summary, the EPA microbiological methods manual fills the specific needs and legal requirements of the Agency to standardize methods in support of its enforcement, compliance monitoring, ambient monitoring and research programs. The methods chosen are the best available, allow limited options and represent the consensus of EPA microbiologists. The manual format allows for rapid response to changing Agency needs, offers opportunity for frequent revisions, guides the analyst in the selection and use of the best methods for his/her purposes and features a stepwise "cook book" form for application at the bench level.

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QUESTION AND ANSWER SESSION

C.W. Hendricks, Office of Drinking Water, U.S. Environmental Protection Agency, Washington, D.C.

When will the manual be available?

R.H. Bordner, Environmental Support Laboratory, U.S. Environmental Protection Agency, Cincinnati, Ohio

The target date for printing is September, 1977. The manual should be available by the end of the calendar year.

S.M. Morrison, Department of Microbiology, Colorado State University, Fort Collins, Colorado

In response to the Drinking Water Law many laboratories are buying equipment and instrumentation and preparing to perform tests they have never done before with technicians who are completely unfamiliar with the tests and equipment. This is particularly true of the smaller laboratories. Have any provisions been made for this problem?

R.H. Bordner

EPA recognizes the need for training as an important part of the program to upgrade the laboratories. Training by the States is certainly encouraged; to our knowledge funding has not been provided by EPA.

S.M. Morrison

I would like to see the use of regional or district laboratories which do have this expertise encouraged.

W. Litsky, Department of Environmental Sciences, University of Massachusetts, Amherst, Massachusetts

You described briefly the relationship of the EPA methods to *Standard Methods*. How do the methods in the EPA manual relate to those being developed by other organizations such as ASTM (the American Society for Testing and Materials)?

R.H. Bordner

EPA members are participating in the activities of ASTM as well as Standard Methods and other methods—standardizing groups. The methods must be reviewed by the EPA Microbiological Methods Steering Committee. Criteria for inclusion of new methods in the manual, in the future state that the method should satisfy the needs of the Agency, show some advantages over present methods, be validated by the developer and other laboratories, be relatively easy to use and practical, etc.

C.W. Hendricks

You mentioned internal review of the manual. Are there provisions for the manual to be reviewed by experts in aquatic microbiology outside the Agency?

R.H. Bordner

We do plan on an external review.

Evaluation of Standard Procedures: Standard Methods

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In 1905, after almost ten years of deliberations and discussions, the American Public Health Association published the first edition of what is now called *Standard Methods for the Examination of Water and Wastewater.* It was a book devoted largely to the enumeration and identification of bacteria in water but it did include methods for chemical analysis as well. The intervening 72 years have seen tremenous expansion of the manual, widespread legal or quasi-legal acceptance of the methods it contains, and a total of fourteen editions. The 15th edition is in preparation and expected to be available by 1978.

Although APHA has continued as the publisher, it was joined by the American Water Works Association in 1925 and in 1947 by the Water Pollution Control Federation in preparing this widely used and widely accepted manual of methods. The three sponsoring societies have different internal procedures for dealing with the administrative and technical details of producing *Standard Methods* but all of these are coordinated by a Joint Editorial Board that consists of the chairman of each society's standard methods committee. Within this framework, APHA has major responsibility for the sections on microbiology.

The current edition includes 131 pages (out of a total of 1193) devoted to the bacteriological examination of water and wastewater. Principal emphasis is given to counting organisms indicative of fecal pollution such as total coliform bacteria, fecal coliforms, and fecal streptococci by either a multiple tube dilution or a membrane filter technique and to enumeration of the total bacterial count. Brief sections are devoted to detection of pathogens such as *Salmonella, Shigella,* enteropathogenic *E. coli,* and leptospires. Tentative procedures are included for viruses in water, *Staphylococcus* and *Pseudomonas aeruginosa* in swimming pools, fungi, yeasts, actinomycetes, and nematodes. A section on the identification of nuisance iron and sulfur bacteria is included also.

The sections devoted to indicator organisms do not emphasize a strict taxonomic approach to identification since that has never been considered of primary importance in public health. Thus, for example, methods for *Escherichia coli* or *Enterobacter aerogenes* are much less important than procedures for fecal coliforms which are defined as coliform bacteria capable of producing gas from lactose at 44.5° C. This difference stresses the pragmatic approach to counting organisms originating in the gut of warmblooded animals rather than specific identifications.

On both a national and an international level in recent years there has been a proliferation of organized interest in standardization of methods in aquatic microbiology. In the United States, as we have heard, the Environmental Protection Agency is in process of preparing a manual of bacteriological methods that will be a companion volume to their already published Methods for Chemical Analysis of Water and Wastes (EPA-625-/6-74-003, 1974) and Biological Field and Laboratory Methods for Measuring the Quality of Surface Waters and Effluents (EPA-670/4-73-001, 1973). It has been argued that EPA as a regulatory agency requires procedures that will be uniformly used throughout the country without permitting either deviations or selections among alternative procedures. However, since EPA leans heavily on *Standard Methods* it may be questioned why a separate manual is at all necessary.

In the face of the long history of *Standard Methods* activities in water bacteriology it is even stranger that the American Society for Testing and Materials, which does publish a manual on water examination, should organize subcommittees of their major water committee, D-19, to consider analyses in water bacteriology and, presumably, ultimately to publish them. These activities raise an interesting point for consideration: Since the number of scientists engaged in any specific aspect of water analysis, in this instance microbiological testing, is reasonably small, would it not be more productive to collaborate and through pooled efforts produce a single, best document of methods rather than to have a proliferation of competing ones that may be of lesser quality and, simply because of their existence, confusing to users?

Making the situation even more complex are the current efforts of the International Standards Organization (ISO) and the European Office of the World Health Organization to produce separate manuals of methods of water analysis. These groups are attempting to reconcile methodological differences among, primarily, European national standards but they too lean heavily on *Standard Methods*.

All of these activities demonstrate two things: first, that there is an urgent need for standardized methods of water analysis so that meaningful and comparable results may be obtained irrespective of the laboratory or country in which the analyses are made and, second, that collaborative rather than competitive action is necessary to insure maximum advantage to all those concerned with water.

In this discussion I have turned from giving an abbreviated report of the microbiological contents of *Standard Methods for the Examination of Water and Wastewater* to making a plea for a united attack on one of society's major problems, water and water pollution control. If we were to examine the changes in health effects of water that have occurred during the 70-year history of *Standard Methods* we would see clearly the disappearance or reduction in enteric waterborne disease and the tremendous impact of the bacteriologist on this improvement. Unfortunately we cannot rest with past victories but must be ever alert to continuing or to newly emerging problems. Unified action by the world's aquatic microbiologists in research, methods development, and water quality control is the key.

Although I have not had an opportunity to review in depth the EPA bacteriological manual, I have seen the table of contents and the pages dealing with coliform analyses. On doing a line-by-line comparison between the EPA manual and *Standard Methods*, I was struck by their similarities in approach and the identical methods included in both. This is not surprising considering *Standard Methods'* long history and the important role in its preparation that has been and is being played by bacteriologists from EPA and its predecessor organizations. With this in mind, I only can conclude that the need for an EPA bacteriological manual is nonexistent. Methodological or other changes desired or required by EPA in its regulatory activities always can be introduced into *Standard Methods* with none of the cost in dollars and high level bacteriologists' time. As an example, it may be argued that *Standard Methods* is deficient in not having an adequate quality control section. Since Messrs. Bordner and Winter, co-editors of the EPA manual, are respectively chairman and member of the *Standard Methods*

Joint Task Group responsible for preparing the section on quality control, the remedy is obvious and immediately at hand.

In conclusion, the Water Supply Program of EPA has available to it the best efforts of bacteriologists, including those from EPA, in providing analytical methods for evaluating microbiology standards. These exist in *Standard Methods for the Examination of Water and Wastewater* which should be adopted as the sole methods source. Exclusion of unacceptable alternative methods can be achieved by appropriate page or section citations in EPA regulations.

Virus Detection

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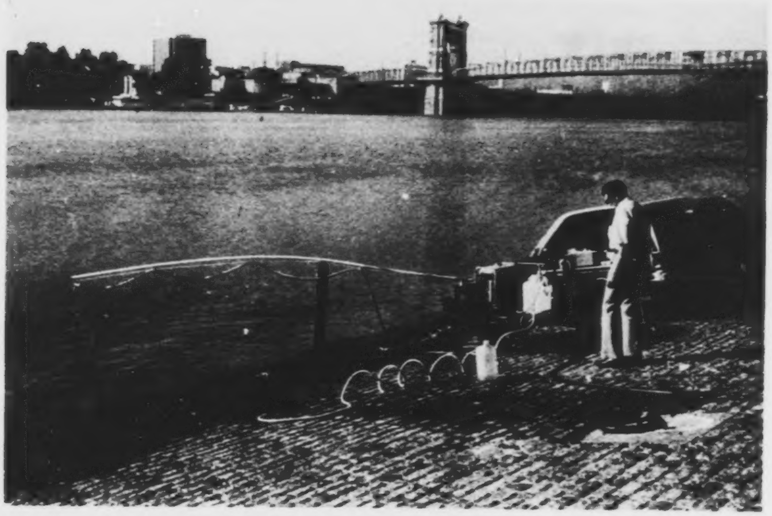

Field Virus Survey Unit (US Environmental Agency)

I have been assigned the topic of "Virus Detection", as it relates to Evaluation of the Microbiology Standards for Drinking Water. I plan to interpret this charge rather liberally.

I shall begin by noting that the purpose of standards is to protect public health, that viruses are a class of pathogens sometimes transmitted by water, and that I have been unable to find the word "virus" in the National Interim Primary Drinking Water Regulations. In the spirit of the Regulations, I had better define the term before I use it further. The viruses share three most important properties:

1. They are transmitted from host to host in the form of discrete particles too small to be seen with a light microscope.
2. They multiply only inside suitable living host cells.
3. They cause mysterious, incurable diseases (in the sense that when your physician tells you, you have a "virus", he is saying he doesn't think he can do anything for you). Fortunately, most individuals, if they are otherwise healthy, recover from most virus infections without any substantive medical assistance.

The only virus disease *proved* to be transmitted currently in water in the United States is hepatitis A, formerly known as infectious hepatitis (11).

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Still, some of the gastroenteritis which results from drinking contaminated water is probably caused by virus (2) and many viruses known to cause other diseases are detected in sewage-polluted waters (10). I don't want to belabor the point, but I think it is worth noting that all of the viruses thought to be transmissible in drinking water are shed in human feces, and that these virus-containing feces have been diluted by a factor of approximately 2000 as they occur in domestic wastewater. Therefore, virtually all threat of virus transmission through drinking water could be eliminated by perfecting and requiring the use of some of the alternatives to common water-flush toilet (also known as the "W.C.").

We can assume that this is unlikely to happen in the near future and that viruses will continue to occur in raw water sources supplying some community systems. I ought, then, to point out that conventional water treatment techniques were not specifically developed to remove or inactivate viruses, and that viruses have a perverse tendency to adhere to each other (5) as well as to suspended solids in water (12). These factors, and the relative resistance of viruses to chemical disinfectants (9), would appear to complicate the task of producing virus-free drinking water from contaminated raw water or from wastewater. Still, known incidents of virus disease associated with drinking water have resulted either from chronic under-treatment of raw water that was known to be contaminated, or from cross-contamination or back-siphonage that introduced virus-containing wastewater into otherwise safe finished water during distribution (4). Rather than viewing virus transmission through water as an exotic problem requiring new and unprecedented countermeasures, I suggest that it be seen as another potential consequence of making some of the same old mistakes. Such lapses are infrequent and are not equally probable: under-treatment has most often been seen to occur in private or semi-public water systems; incidents involving community water supplies have most frequently entailed recontamination of the finished water during distribution (4).

One would like to have some other means than human disease to detect the presence of virus in drinking water. The task of recovering virus is complicated by the extremely small quantity of virus material that is likely to be present in a water sample and by the tendency of the virus particles to adsorb to suspended solids (13). However, quite elegant methods have been devised for recovering virus particles from samples as large as 1900 liters (~ 500 gallons) or greater. The virus particles in the concentrated sample extract have still to be detected by the infections they may produce in tissue cultures (3). Such infections become perceptible only slowly: inoculated tissue cultures must usually be incubated and observed for days or weeks. The potential for success varies: no tissue culture is known to support replication of the hepatitis A virus, but tissue cultures are probably more sensitive than the host's body to some of the other intestinal viruses which might occur in water (Cliver, unpublished data). Although, there will always be room for improvement in such techniques, we have now reached the point where a "Tentative Standard Method" for detecting viruses in water exists (3). The method has been shown to recover experimentally-inoculated viruses efficiently from large-volume samples of water (6). No virus has been detected in approximately 175 large-volume samples of finished community drinking water by this method, even though the raw water sources of some of these communities were known to be highly contaminated (1; E.W. Akin, EPA Cincinnati, personal communication).

The most obvious explanation for these negative test results is that virus was absent from the samples, although other reasons can be envisioned.

Because it is difficult to detect viruses in water, it is reasonable to seek "indicators" (organisms or substances whose presence in water is likely to accompany that of virus). In fact, there are many possible indicator systems: Coliforms, fecal coliforms, and fecal streptococci suggest fecal contamination of water; however, they differ from viruses in the attrition they are likely to undergo in the environment or during water treatment. Turbidity, standard plate count, and free residual chlorine (or lack of it) have, at best, an indirect association with the likelihood that virus has contaminated water. Coliphages (bacteriophages specifically infectious for *Escherichia coli*) as a group are indicative of fecal contamination of water and *may* lose their infectivity at a rate comparable to animal viruses (8); however, there seems to be some problem in selecting a "best" host strain of *E. coli* in which to detect the coliphages (14).

Finally, the vaccine strains of the human polioviruses have been proposed as indicators because they are almost always present in urban sewage, are likely to lose infectivity at approximately the same rates as many other intestinal viruses, and may be more rapidly detectable than viruses in general, using a fluorescent antibody test procedure (7). Unfortunately, polioviruses are not *always* present in sewage, and the detection methods are a good deal more complex and exacting than most Standard Methods microbiological procedures.

By now, you are probably wondering how it is ever possible to determine that a community's drinking water does not contain virus. In fact, it is not possible to demonstrate the complete absence of virus from each day's total production of finished water. The most direct test of the safety of drinking water is drinking the water. However, given the two-liter-per-day consumption figure upon which the Maximum Contaminant Levels are supposed to be based, only about 1% of the total water used at home (and, therefore, even less of the daily per capita total water consumption) is drunk. The rest is used in a variety of ways that are unlikely to allow manifestation of any virus it contains.

The water could also be tested for virus by the procedures mentioned previously. I believe that the surveys now in progress represent the most proper use of the Tentative Standard Method and its counterparts—these are not procedures appropriate for routine quality control of finished waters by individual suppliers; neither can they reasonably serve as the basis for establishing or enforcing Maximum Contaminant Levels for viruses. What the survey results seem to show is that virus-free water is attainable by means of the best current treatment methods, if these are conscientiously applied, even when raw water sources are far from ideal. Less rigorous treatment is likely to be necessary with high-quality, protected raw water sources. The evidence indicates that the means to produce virus-free finished water are at hand, at least for raw waters that do not consist principally of sewage effluent. The most important task at this time is to ensure that these means are employed, absoluely without fail, by water suppliers. Whether virus testing involves one "large" sample or several small ones per day, only a minute portion of a community's daily water output could be monitored in this way, at a cost in resources that could better be applied to other surveillance tasks. The findings of the EPA community water sutvey indicate that the results of such testing are likely to be negative, but this obviously does not guarantee that the consumer is receiving virus-free drinking water.

I believe that the most consumer protection that can be obtained for the consumer's dollar will derive from means already at hand. Suppliers should be helped in selecting treatments appropriate to the quality of their raw water. For any source other than extremely well-protected ground water, treatment should probably include at least the following: 1) coagulation and sedimentation, 2) filtration, and 3) chemical disinfection (I am not now prepared to recommend treatments for direct reclamation of urban effluents). The physical treatments which are suggested should serve to remove protozoan cysts, a very real cause for concern, as well as most solids that might protect virus from chemical disinfection. Beyond selection and adaptation of treatments appropriate to the quality of the raw water, adequate consumer protection requires that the supplier by supervised to ensure that the treatments are properly performed all of the time. Microbiological testing of the finished product alone won't do the job. There must be monitoring of both microorganisms and turbidity, as well as a continuous record of the processing as it is done. Then the quality of the finished water must be sustained all the way to the user's home. The presence of a free chlorine residual, or the absence of coliform organisms, at the tap is reasonable evidence that cross-contamination or back-siphonage has not occurred to degrade the finished water during distribution.

I believe that this discussion supports a few conclusions: First, it is reasonable to test for viruses in finished drinking water on occasion, but it is not reasonable to expect drinking water suppliers to do it. Second, Maximum Contaminant Levels (as defined in the Safe Drinking Water Act of 1974), are not appropriate for viruses. Finally, the basis for protecting the public from water-borne virus disease lies rather in detecting lapses in the practices of treatment and distribution than in detecting the virus in the finished product.

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Disinfectant Resistant Organisms *

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Much of the dialogue at this symposium has been based on laboratory disinfection studies in which the die off rates of standard strains of viruses have been measured as a function of time at various levels of chlorine residuals. While this specific information is technically sound, it overlooks the variations in resistance to chlorine inactivation among various strains of viruses or among individual members of a specific virus strain. Thus, it becomes questionable to measure a chlorine residual and assume that this will assure a given level of disinfection of all viruses. Any indirect standard must be viewed with some misgivings. If a water is to be assured to be free of a particular organism, that organism must be monitored even if infrequently.

Virus isolates recovered from well-treated finished water (2) have been questioned, to no small extent on the basis that they were recovered from a water of very low turbidity in the presence of chlorine residuals in excess of one milligram per liter (1). We have examined isolates from one such study in considerable detail and are convinced that they are sufficiently different from any available laboratory strains to assure that they are natural isolates and not artifacts resulting from contamination (3).

Most recently, two of these isolates were grown to high titers, carefully purified by sedimentation in an ultracentrifuge, and subjected to chlorine inactivation. Their inactivation rates were compared with those of two standard poliovirus 1 strains (LSc and Mahoney) grown up and purified in the same way. They showed a chlorine inactivation rate orders of magnitude lower than that of either of the two standard controls. These data shown, in part below, will be published in detail shortly.

VIRUS STRAIN	*LOG SURVIVAL FRACTION (30 MINUTES)*	*FREE CHLORINE RESIDUAL (mg/l)*
Mahoney	−4.6	0.39
LSc	−7.79	1.34
Isolate 1	−1.74	0.92
Isolate 2	−3.83	1.31

The point to be noted is that all viruses, in fact all strains, or even all members within a single standard strain, do not respond in the same way when subjected to disinfection conditions typical of those applied in a well operated drinking water plant. To assume that all viruses will exhibit the same die off rate as that shown by standard laboratory strains is not technically sound. Therefore, to replace a direct standard, whether it be coliform or virus, with an indirect standard of turbidity and chlorine residual can not be defended on the basis of experimental evidence.

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* This paper was submitted after the Conference.

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The Relationship of Turbidity to Disinfection of Potable Water

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Introduction

Although turbidity has been used as a measure of potable water quality for many years, increased attention has been focused on this parameter as a result of its inclusion at a Maximum Contaminant Level (MCL) of 1 Nephelometric Turbidity Unit (NTU) in the National Interim Primary Drinking Water Regulations. Turbidity in drinking water can be caused by a large variety of particles which can originate from many sources. Turbidity may be present as a result of incomplete removal by treatment processes of particles present in the raw water source or may result from problems with water treatment techniques such as coagulation. Turbidity may also originate within the distribution system as a result of corrosion, growth of microorganisms, line repairs, etc.

Because of the current emphasis on turbidity in potable water, the significance of turbidity, particularly as it relates to disinfection efficiency during water treatment, will be discussed from the historical point of view as well as in the light of recently developed information. Problems associated with the use of turbidity as a contaminant limit and some possible alternatives for future consideration also will be discussed.

Turbidity as a Potable Water Standard

Turbidity in water is caused by the presence of suspended matter such as clay, silt, finely divided organic and inorganic matter and microscopic microorganisms. Turbidity is defined and expressed as an optical property of water and is measured by determining the degree of light scattering by the particulates present in the sample (1). Because of the nature of the measurement, turbidity determinations cannot be related either to the quantity (mg/1) or characeristics (size, shape, chemical nature) of the particles present in the sample. Another problem is that different types of turbidimeters, after being standardized on the same turbidity standard, will usually give readings differing by as much as 500% when used to measure the turbidity of a sample containing another turbidity causing substance (12).

Despite these limitations, turbidity has been found to be a useful parameter for measuring the quality of public water supplies. Turbidity first became a part of the USPHS Drinking Water Standards in 1942 when a tolerance limit of 10 units was established. In 1946 this was changed to a tolerance limit of 10 for filtered water only and a recommended limit of 10 for other waters. In 1962, the recommended limit was reduced to 5 and the tolerance limit was dropped (2). The most recent change embodied in the National Interim Primary Drinking Water Regulations establishes a turbidity Maximum Contaminant Level (MCL) of 1 with up to 5 units allowed if the supplier can demonstrate that the higher limit does not do any of the following:

1. Interfere with disinfection.

2. Prevent maintenance of effective disinfection throughout the distribution system.
3. Interfere with microbiological determinations.

Of the three, the most critical and the one which has generated most concern is the possible interference of turbidity with disinfection efficiency. The second criterion relates to complex problems of turbidity as a source of nutrients and available surface for growth of microorganisms in distribution lines as well as the more direct relationship between turbidity and disinfectant demand. Significance of the third criterion has been shown at high turbidities but an effect in the turbidity range of 1 to 5 has not been clearly demonstrated.

Theoretical Aspects of the Relationship Between Turbidity and Disinfection

For disinfection to be effective, there must be contact between the agent and the organisms that the disinfectant is to kill. Some of the particles which make up part of the turbidity may be particle-microorganism complexes in which a microorganism is adsorbed onto a larger particle. In other cases one or more smaller particles may be adsorbed onto a microorganism or a particle may completely encase a microorganism. In each of these cases varying degrees of protection of the microorganism from the disinfectant may be expected.

The microorganisms of concern in this respect include pathogenic protozoa, bacteria and viruses. Several characteristics of these groups of microorganisms must be considered relative to possible effects of turbidity on their inactivation. One of these is size. The sizes of representatives of these three groups differ by a factor of over 100X (poliovirus 0.03 μm diameter, *E. coli* 0.3 × 1-5 μm, *E. histolytica* cysts 5-20 μm). This would indicate that in the case of virus protection by turbidity, contact between the disinfectant and the microorganism could be prevented by much smaller complexes than would be needed for protection of enterobacteria or protozoans. Size is also important in relationship to the significance of surface charge phenomena in causing aggregation or adsorption of particles. These factors become increasingly important as size decreases and are very important with regard to particles in the size range of enteroviruses. Another factor of importance is the relative innate disinfection resistance of the microorganisms themselves. In general, the enterobacteria are the most sensitive to disinfectants, followed by the enteroviruses, while the protozoans exhibit the greatest resistance (19).

Based on these factors, several assumptions regarding turbidity effects can be made. In order to protect an enterovirus from disinfectant action, a particle, whether produced by adsorption of many extremely small particles onto one virus particle or from dissolution of fecal or other material to form small particles some of which may completely enclose a virus particle, must be larger than 0.03 μm in diameter. Particles which will protect bacteria or cysts must be correspondingly larger. Thus, the particles present in water which are of concern because of possible disinfection efficiency effects are those ranging upwards from 0.03 μm.

The extensive body of literature on the physio-chemical mechanisms involved in virus adsorption onto surfaces and the influence of these interactions on the fate of viruses in the environment was recently reviewed by Britton (5). The adsorption phenomenon is used for virus removal in various waste water and water treatment processes (coagulation, flocculation, filtration and also is applied in methods developed for concentration and detection of low levels of virus from large volumes of water. Because of the way in which viruses, particularly enteroviruses, are produced, excreted, and transported (intestinal tract → feces → water) and the number and variety of particles present in natural waters it is likely that many enteroviruses exist in the environment as adsorbed, rather than free, particles (4).

Results of Research on Protective Effects of Turbidity

Although the concept that certain particles present in water may protect microorganisms from inactivation by disinfection certainly is logical, little direct evidence of such protection has been demonstrated. Clarke and Chang (9) stated that turbidity was responsible for disinfection failure which resulted in the Delhi infectious hepatitis outbreak. An AWWA committee (3) stated that "Effective disinfection can be carried out only on water free from suspended material." Culp (11) stated that for good viral disinfection turbidities of 0.1 JTU were preferable. Cookson (10) stated that the ability of disinfection to produce virus-free water depends on removal of turbidity to no more than 1 JTU.

A few studies have provided evidence that the presence of particulate matter interferes with effective inactivation of viruses and bacteria in water. In an early rather crude study, Neefe *et al* (15) showed that feces from an infectious hepatitis patient when suspended in water caused hepatitis in two of five human volunteers after exposure to a chlorine residual of 1.1 mg/l for 30 minutes. In a similar experiment, when the water was first coagulated and then filtered before disinfection to the same chlorine residual, none of five volunteers contracted hepatitis. Change *et al* (7) demonstrated that enteric bacteria and enteroviruses ingested by aquatic nematodes were completely protected against high residual chlorine levels (80-90 mg/l) even when more than 90% of the nematodes were immobilized. Tracy *et al* (18) reported the persistence of coliforms in drinking water which was highly chlorinated. The problem was most severe when turbidities were between 5 and 10 units. They concluded that the most likely explanation was that the coliforms had been ingested by *Crustacea* present in the water and were thus protected from contact with the disinfectant.

In a discussion included as a part of a paper by Clarke *et al* (8), W. W. Sanderson and S. Kelly of the New York State Health Department, Division of Laboratories presented evidence that turbidity interfered with disinfection in a water supply. They studied an impounded water supply receiving no treatment other than chlorination. The concentration of free chlorine residual in samples from household taps after a minimum of 30 minutes contact time varied from 0.1 to 0.5 mg/l and the total chlorine residual was between 0.7 and 1 mg/l. These samples consistently yielded confirmed coliform organisms. Turbidities in these samples varied from 4 to 84 TU's and microscopic examination showed iron rust and plankton to be present. They concluded ". . . coliform bacteria were imbedded in particles of turbidity and were probably never in contact with the active agent. Viruses, being smaller than bacteria, are much more likely to escape the action of chlorine in a natural water. Thus, it would be essential to treat water by coagulation and filtration to nearly zero turbidity if chlorination is to be effective as a viricidal process."

Results of our research on this problem as well as the results of other recent research provide more definitive information regarding the relationship of low levels of turbidity and disinfection. Our research in this area has been designed to determine whether significant differences in disinfection

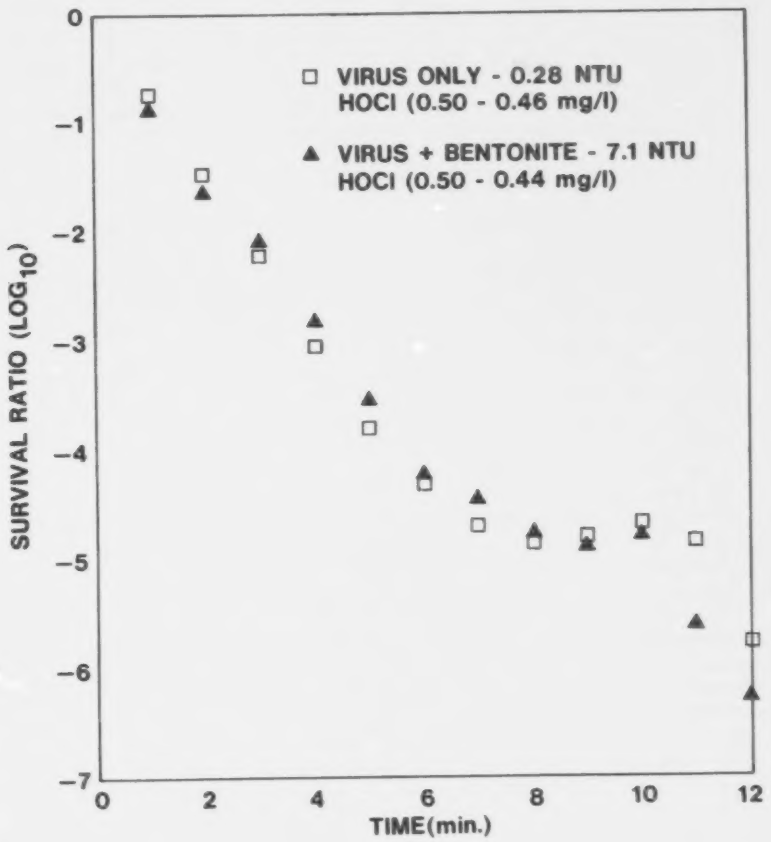

Figure 1. INACTIVATION OF BENTONITE - ASSOCIATED POLIOVIRUS 1 BY CHLORINE (pH 6.0, 5° C).

efficiency can be specifically related to turbidities of 5 NTU and 1 NTU. All experiments were conducted at 5°C and pH 6.0. Under these conditions virtually 100% of the free chlorine residual is present as hypochlorous acid (HOCl) which is the most effective chlorine species for disinfection. The LSc2ab strain of attenuated poliovirus type 1, treated to remove chlorine demand, was used.

Studies of effects of inorganic turbidity on disinfection were done using virus adsorbed on bentonite clay and virus precipitated with aluminum phosphate ($AlPO_4$). Volumes of virus associated with bentonite or $AlPO_4$ sufficient to produce a turbidity of ~ 5 NTU were added to stirred reactors containing 0.5 mg/l HOCl. Centrifugation results showed that 65-80% of the bentonite-virus mixture was bentonite associated and over 90% of the $AlPO_4$-virus mixture was $AlPO_4$ associated. An example of results using

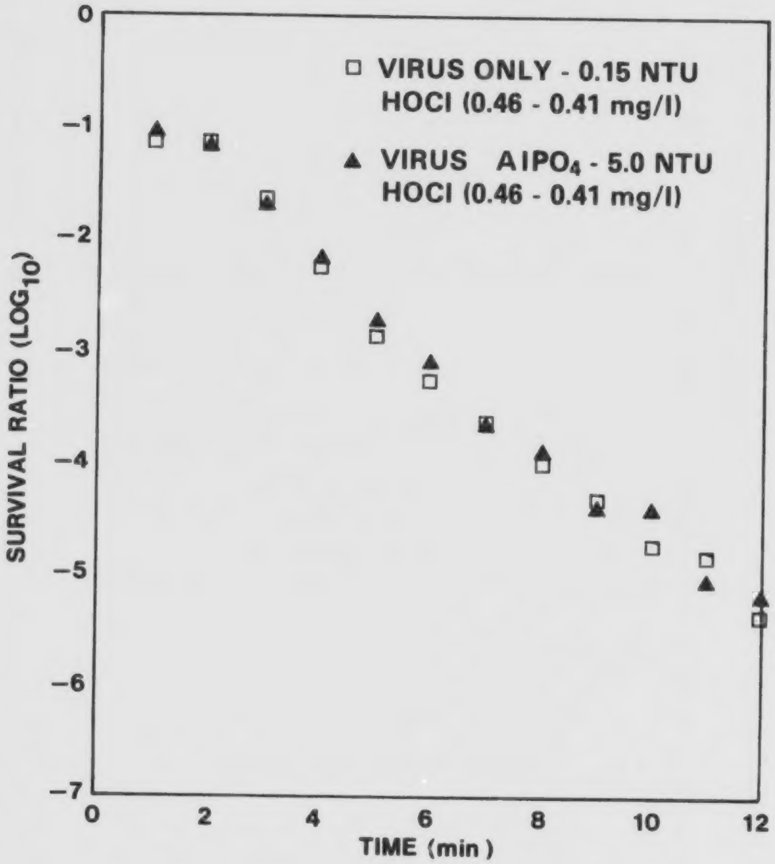

Figure 2. INACTIVATION OF $AlPO_4$ - ASSOCIATED POLIOVIRUS 1 BY CHLORINE (pH 6.0,5°C).

bentonite associated virus are shown in Figure 1. The results show that the disinfection curves are very similar with no indication of a protective effect. Results using $AlPO_4$ associated virus are shown in Figure 2. In this case also, inactivation rates were very similar with no indication of a protective effect.

Studies of effects of organic turbidity on disinfection were done using cell associated virus. Cell associated virus consisted of virus-cell debris complexes formed by virus infection and destruction of cell cultures. The cell debris was harvested and processed by repeated washing and low speed centrifugation to remove extraneous chlorine demand. The preparations, when diluted to a turbidity of ∼ 1 NTU, contained virus levels of approximately 1×10^6 PFU/ml. Preparations at 1 NTU showed some chlorine demand associated with the cell debris. Therefore, experiments using these preparations and comparative studies with clay associated, $AlPO_4$ associated and free virus were

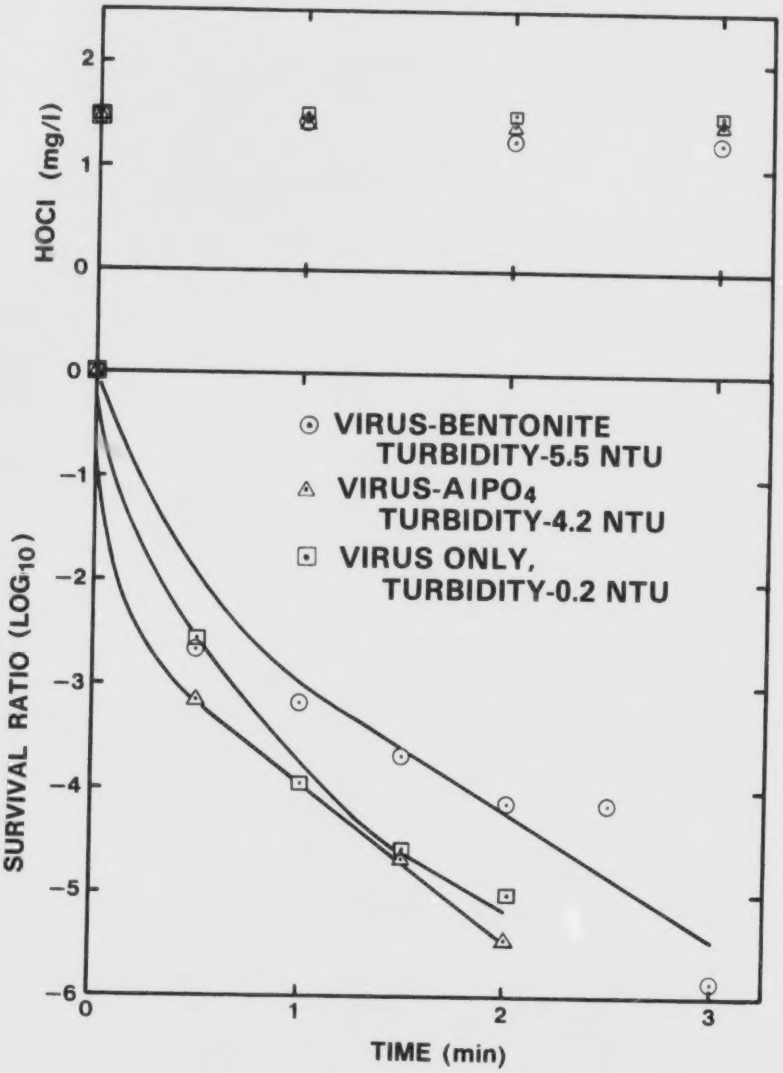

Figure 3. INACTIVATION OF POLIVIRUS ASSOCIATED WITH $AlPO_4$ AND BENTONITE BY 1.5 mg/1 HOCl.

conducted at higher HOCl levels (1.5 to 3.0 mg/l). Results of an experiment comparing inactivation of clay associated, $AlPO_4$ associated, and free virus at 1.5 mg/l HOCl are shown in Figure 3. Inactivation rates were very similar, with bentonite associated virus showing a slightly lower rate of inactivation than $AlPO_4$ associated or free virus. Figure 4 shows a com-

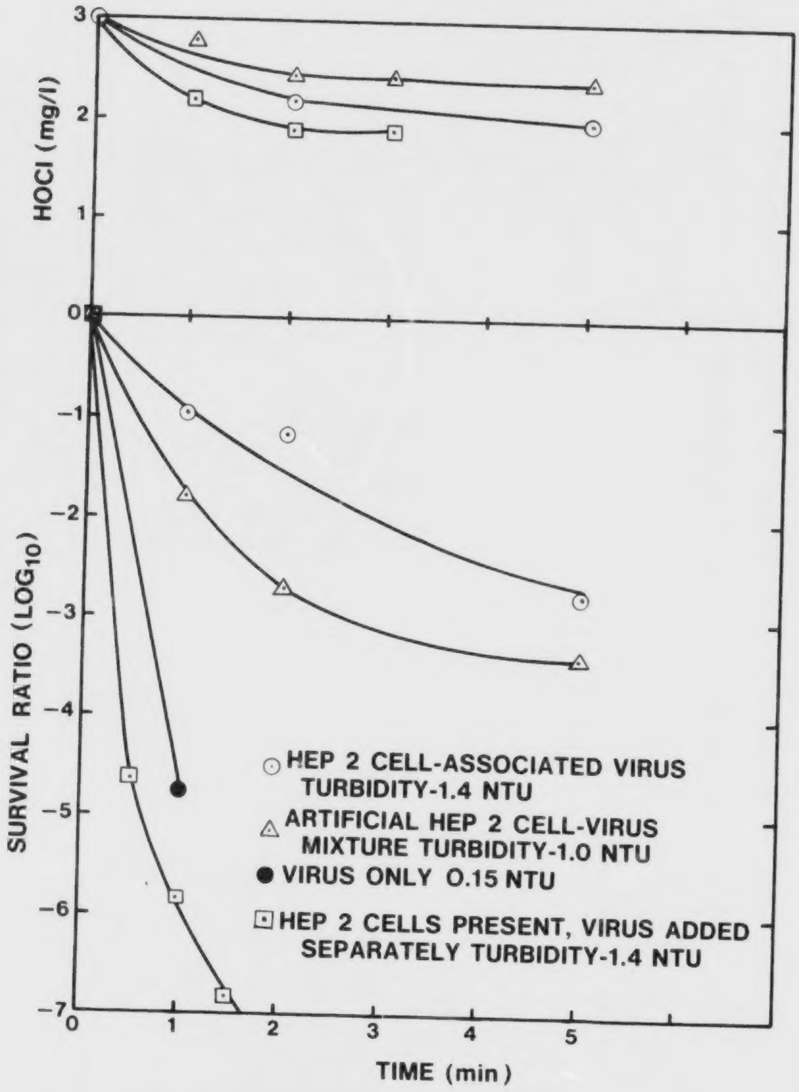

Figure 4. EFFECT OF CELLULAR MATERIAL ON RATE OF POLIVIRUS INACTIVATION BY CHLORINE.

parison of disinfection rates of free virus, cell-associated virus and an artificial cell-associated virus mixture prepared by mixing virus with cells which had been harvested, washed, and stored at 5°C for 1-2 days. The artificial mixture was stored overnite at 5°C before use. The results show a pronounced protective effect of virus associated with cells. Also shown in

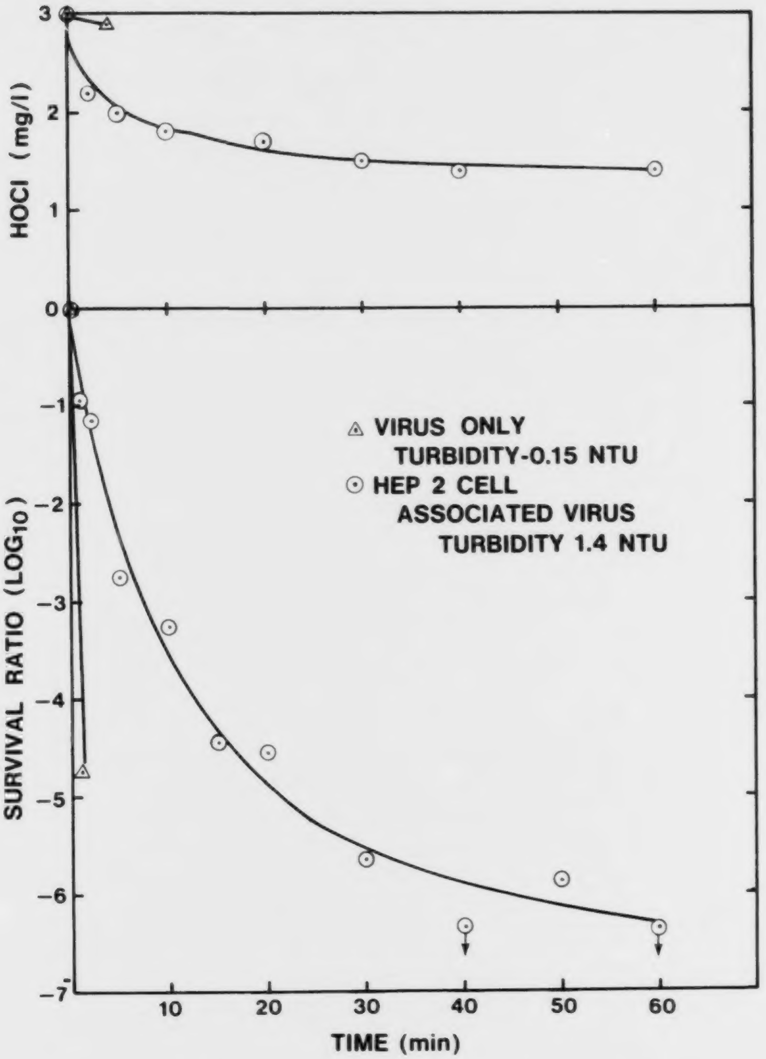

Figure 5. INACTIVATION OF CELL - ASSOCIATED POLIVIRUS BY CHLORINE (60 min.).

Figure 4 is an inactivation curve in which washed cells were added to the disinfection reactor followed immediately by virus. This simulated conditions in which virus inactivation was occurring in the presênce of chlorine demand created by the cellular material. The results show that the presence of chlorine demand had no effect on the inactivation of the virus. Figure 5 shows the results of a longer term experiment on inactivation of cell associated virus.

HOCl levels declined from an initial level of 3.0 mg/l to 1.5 mg/l, most of the decline occurring in the first 5 minutes of the experiment. Even though the turdidity was only 1.4 NTU, virus was detected even after 50 minutes exposure to 1.5 mg/l HOCl. Free virus exposed to 3.0 mg/l HOCl was reduced more than 5 logs in less than two minutes.

In two other recent studies, the effect of inorganic particles on virus disinfection has been studied. Boardman (6) reported that inorganic particles including alum, calcium carbonate and kaolin did not interfere with bacteriophage T7 inactivation by chlorine but did interfere with the rate of inactivation of poliovirus. The poliovirus results are shown in Table 1. The experiments were conducted at 22.5°C at a residual chlorine level of 0.21 mg/l. The chlorine residual determinations indicate that most of the residual was present as combined rather than free chlorine. Although turbidity was not measured, the amounts of alum (20 mg/l) and kaolin (50 mg/l) were similar to the levels of $AlPO_4$ (50 mg/l) and bentonite (35 mg/l) used in our research. Boardman related the protection shown to extensive virus aggregation enhanced by the presence of particulates serving as foci for virus adsorption. Experiments designed to show this were inconclusive. Boardman also noted that the protective effects were greatest in systems in which information of particulates occurred in the presence of the virus ($CACO_3$ & alum).

Stagg *et al* (17) compared the inactivation by HOCl of bacteriophage MS-2 associated with bentonite clay and in the freely suspended state. Both freely suspended and clay associated phages were inactivated rapidly but the rate of inactivation of clay associated phage was retarded. At equivalent HOCl concentrations ranging from 0.02 to 0.6 mg/l, approximately twice the length of time was required for equivalent inactivation (99%) of clay-associated phages as for freely suspended phages. Reaction rates in both cases were essentially first-order. Time required for 99% inactivation of freely suspended phage was 10 seconds and for clay associated phage 19 seconds at 0.5 mg/l HOCl at 22°C. Turbidity levels for the experiments were in the range of 2 to 4 Jackson Turbidity Units (JTU).

Use of cell associated virus in studies of disinfection of combined sewer overflows by chlorine and chlorine dioxide was also reported by Moffa and Smith (13). Cell associated virus, when mixed with simulated combined sewer overflow water was much more resistant to inactivation by ClO_2 than was

TABLE 1. EFFECT OF VIRUS ADSORPTION ON POLIOVIRUS INACTIVATION BY 0.21 mg/l RESIDUAL CHLORINE[1] AT 22.5°C.

Preparation	pH	Percent Inactivation in 10 Minutes
Polio Only	7.0	99.7
Polio Only	10.1	97.3
Polio + Kaolin (50 mg/l)	7.0	95.8
Polio + Alum (20 mg/l)	7.0	78.4
Polio + Ca CO_3 (600 mg/l)	10.1	84.7

[1] free residual range—0.02-0.08 mg/l
combined residual range—0.12-0.14 mg/l
From: Boardman (6)

freely suspended virus. The results are shown in Figures 6 and 7. Turbidity and disinfection demand characteristics of the water were not given. However, since the cell associated virus was in simulated combined sewer overflow water, it is likely that a high chlorine demand was present.

More recently, we have initiated some studies on the effects of turbidity in the range of 1 to 5 NTU on coliform inactivation by chlorine. These experiments also were conducted at pH 6.0, 5.0°C in 0.05 M phosphate buffer. The coliforms used in these experiments were naturally occurring coliforms associated with solids in unchlorinated primary sewage effluent. The effluent was collected and centrifuged at 2,000 RPM for 15 minutes to sediment the solids. The sedimented solids were washed to remove soluble chlorine demanding substances by repeated centrifugation and resuspension in 0.05 M phosphate buffer, pH 6.0. The concentrated washed solids, when diluted to produce turbidities of 1 and 5 NTU contained coliform concentrations of $1 \times 10^5 - 1 \times 10^6$/100 ml. Inactivation of the coliforms associated with the solids was studied in stirred batch reactors containing 0.5 mg/l HOCl. Coliform levels were determined by standard coliform MPN tests. Completed tests were conducted on all positive tubes. Inactivation of a washed culture of *E. coli* (ATCC 11229) by the same level of HOCl was also determined.

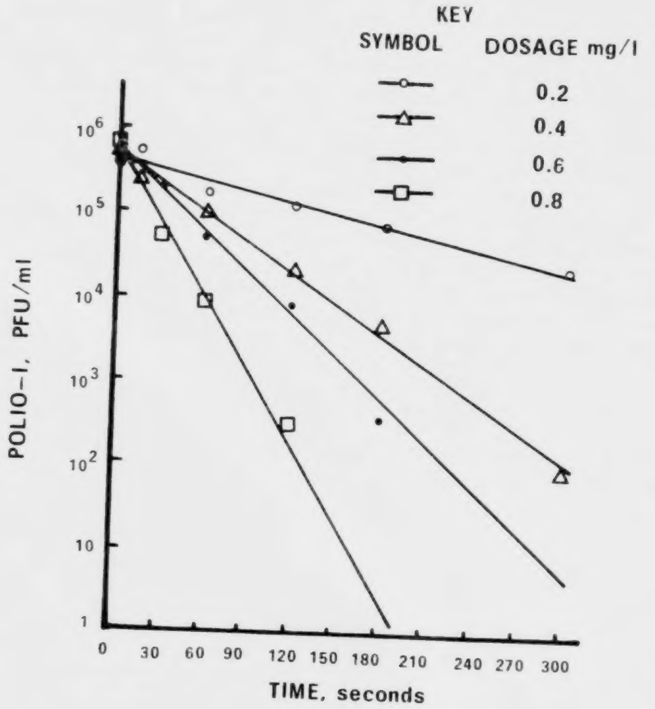

Figure 6. SINGLE-STAGE DISINFECTION OF POLIVIRUS 1 WITH ClO_2 IN A NO-DEMAND SYSTEM. (ADAPTED FROM: MOFFA, 13)

Typical results are shown in Figure 8. It is evident that the naturally occurring coliforms associated with effluent solids were much more resistant to chlorine than was the washed *E. coli* culture. At both 1 and 5 NTU's survival ratios were similar, with coliforms still detectable after 60 minutes exposure to 0.5 mg/l HOCl.

The results of the recent virus studies described above indicate that virus associated with inorganic particles such as clays or $AlPO_4$ is inactivated by HOCl at rates similar or perhaps slightly lower than freely suspended virus. The rates in both cases are essentially first order with no evidence of an extremely resistant fraction. In contrast, disinfection rates of cell associated viruses were not first order, with inactivation rates becoming slower as time of exposure increased. This indicates that various degrees of protection existed with a few complexes being extremely resistant to disinfection.

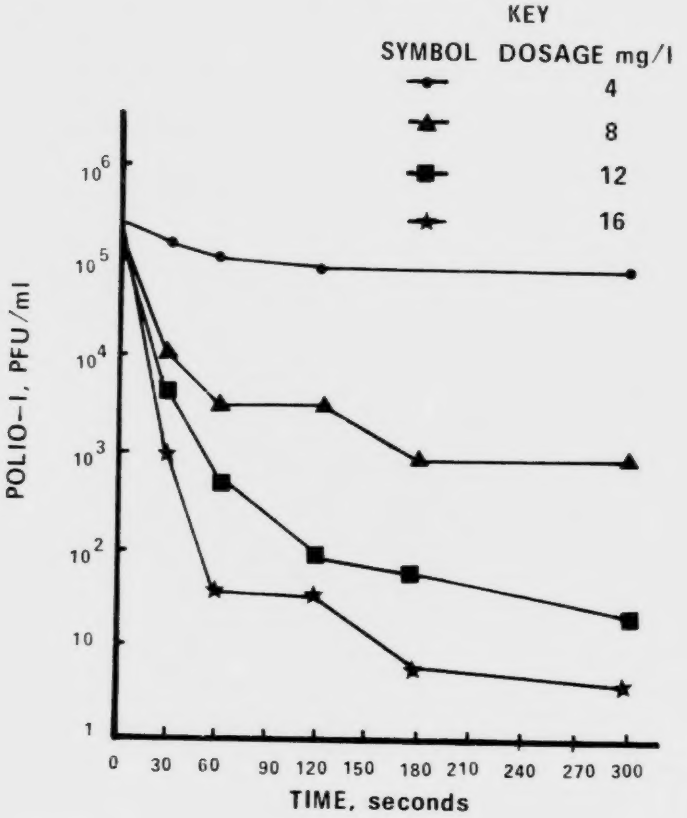

Figure 7. EFFECT OF ClO_2 ON CELL-ASSOCIATED POLIVIRUS 1. (ADAPTED FROM: MOFFA, 13)

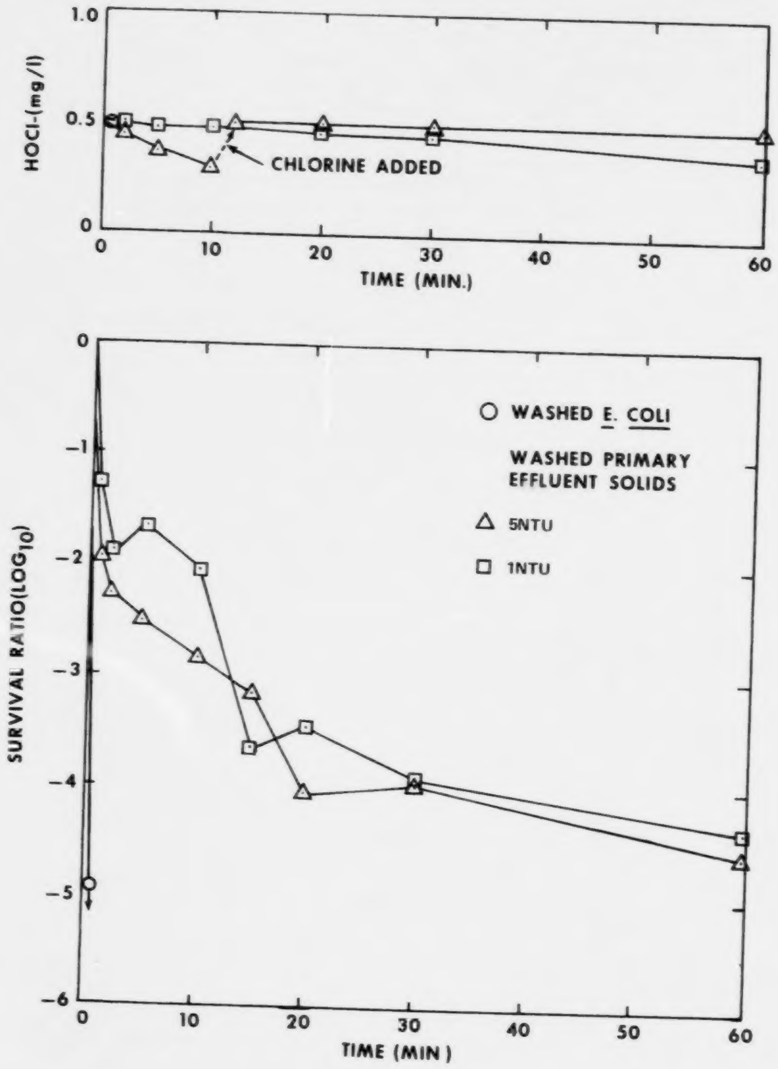

Figure 8. INACTIVATION OF COLIFORMS ASSOCIATED WITH PRIMARY EFFLUENT SOLIDS BY CHLORINE (pH 6.0, 5.0°C).

Discussion

In these studies, the measure of the presence or absence of viable virions after exposure to the disinfectant was the ability of the virus-particulate complex to initiate infection of a cell, thereby resulting in a visible plaque. Recent studies have shown that enteroviruses associated with solids remain infective for cell cultures (14) and for animals (16). The mechanism by which virus-particulate complexes initiate infection is not known. It is possible that the complex is broken at the cell surface and the virion enters the cell as an unadsorbed particle or that the entire complex enters the cell. It is known that various chemicals, notably serum protein can prevent adsorption of virus to inorganic particles and also can elute virus previously adsorbed to such particles. Because of the nature of the assay system (living cells) it is inevitable that some soluble protein will be present at the cell surface. Because of this, it is difficult to determine the details of cell infection by adsorbed virus. It is also possible that viable virions, completely encased in feces, cell debris, etc. and thereby completely protected from disinfection, may exist and remain undetected. In a few instances in the studies we conducted, efforts were made to release protected cell associated virus by freezing and thawing of samples collected at various intervals during disinfection. The results of these studies were equivocal. Because of the complex digestive processes which occur following ingestion, such virus could be freed from encasement and being viable, initiate infection in the host. Thus, virus which remains viable but undetectable in cell culture systems could possibly be detectable in an animal assay system.

While we do not know how well cell associated viruses simulate viruses as they exist in natural waters, it is likely that this model is more representative of natural conditions than the model employing freely suspended viruses. The results of the disinfection studies of naturally occurring solids associated coliforms show inactivation curves similar to those shown by cell associated viruses. This would indicate that similar effects would be seen with naturally occurring viruses present in such solids.

In interpreting the results of these studies, particularly the virus disinfection research, it should be kept in mind that the protective effects shown, while they well may reflect what actually occurs during water treatment, represent a tremendous magnification of the virus levels which might be present under "real world" conditions. Whereas in laboratory experiments, we can begin studies with tremendous numbers of viruses ($\sim 10^6$ PFU/ml), actual numbers present in some of the worst raw waters used as potable water sources may contain on the order of 1 PFU/gallon.

The information presented above has been limited to the consideration of turbidity interferences with disinfection of potable water. Several other very important aspects of turbidity as related to health hazards associated with potable water have not been addressed. These include the use of turbidity as an indicator of the presence of particulates such as asbestos which may pose a chemical hazard or as an indicator of the presence of microbiological forms such as *E. histolytica*, bacterial spores, and perhaps *Giardia lamblia* which have or are believed to have a high innate resistance to disinfection. These aspects must be taken into account in considering requests for allowing turbidity limits higher than 1 NTU.

Conclusions

As stated previously, turbidity measurement is well established as a useful operational guide for assessing the performance of various processes such as coagulation, sedimentation and filtration in water treatment plant operation. Unfortunately because of its lack of specificity in determining the nature of the turbidity particles present, turbidity does not appear to be the ideal indicator of disinfectability of potable water since the interference of turbidity with disinfection depends much more on the types of turbidity present than the number of turbidity units present. The bacteriological data derived from the primary effluent studies indicate similar disinfection curves at turbitities of 5 and 1. Based on this, one could assume that water treatment processes which reduce this type of turbidity to 1 NTU rather than 5 NTU would result in an approximate 5 fold increase in safety factor. In other words the disinfectant resistant portion would still be present but at a 5 fold lower concentration. Although cell-associated virus studies were not conducted at both turbidities, it seems reasonable that the same observation could be made in the case of turbidity caused by clays and $AlPO_4$. Interference with disinfection efficiency is either negligible or relatively minor. It is likely that little in the way of increased safety factor would be gained from reducing turbidity caused by these types of particles from 5 NTU to 1 NTU.

While it is evident that turbidity is at present the established parameter, future consideration should be given to other parameters as a possible measure of particulate interference with disinfection for use as regulatory criteria. Parameters such as total organic carbon or nitrogen in the suspended solids present in a particular water might be used to determine whether the solids are organic or inorganic. Determination of chlorine demand of the solids present in a particular water might also be used to distinguish inorganic from organic particles.

Further consideration of the relevance of coliforms as indicators of viral safety of disinfected water may also be in order. The results of the studies on inactivation of naturally occurring coliforms associated with primary effluent solids indicate that such coliforms are far more resistant than freely suspended coliforms. Thus, the coliform group may be a more rigorous disinfection indicator than is indicated by disinfection studies of pure cultures.

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Wastewater Reuse
and the
Problems of Water-Borne Disease

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Introduction

Much has been written about wastewater reuse, a topic that seems to become more popular with time. Since there are many types of reuse applications made of wastewater, perhaps the first thing we should do is define the type of reuse that concerns this presentation: direct, potable reuse. But this still isn't as sharp a definition as I'd like. Webster (10) says that potable means "suitable for drinking." I personally believe that there may be other uses of water that are more demanding than just drinking (11). For example, a water could conceivably not present any trouble to a person drinking it if it contained a substantial population of *Staphylococcus aureus*, but prepare a salad with it and allow it to incubate a few hours and the results could be quite different. Hence, I usually generalize the type of reuse that I have in mind as: for intimate human usage.

Another definition is essential to our discussion today. Webster (10) defines disinfection as "the freeing from infection esp. by destroying harmful microorganisms." The Glossary of Water and Wastewater Control Engineering (3) defines it as "the art of killing the larger portion of microorganisms in or on a substance with the probability that all pathogenic bacteria are killed by the agent used." The Glossary definition thus:

1) introduces the concept of disinfection being a rate function—which it is
2) introduces the concept of probability, which is also proper, and
3) excludes viruses.

When applied to drinking water, the Glossary definition must be updated to include pathogenic viruses. Until it is, we must be careful that we define the interpretation of disinfection we are talking about.

Organisms of Concern

Since the wastewater reuse under consideration concerns the most intimate human usages, and since wastewater can conceivably include anything, we must address virtually all pathogenic agents. It would be inordinately naive to be concerned only with enteric organisms even though these are expected to be the most abundant.

From a disinfection point of view, the organisms of concern can be grouped as follows:

- Bacteria
  - Vegetative
    - Encapsulated
    - Non-encapsulated
  - Spores
- Fungi & Yeasts
- Rickettsia

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resistant to all three (Sagik, B. Telephone communication, Feb. 8, 1977).

Some of us in Texas look at our long border with Mexico with some trepidation because we know how readily available antibiotics are across that border, we know that is a large movement of people across that border, and we read periodically in *Morbidity and Mortality*—and in *Science* (4)—increasingly frequent accounts of illness caused by antibiotic resistant strains of bacteria—many of which occur across that border.

2) Viruses

I think that public health engineers, in general, are fairly comfortable with the fact that viruses can be inactivated by HOCl. Now and then we see an article that questions our faith (7). The most recent such occurrences have been discussed by Akin and Jakubowski (1).

The question of virus-presence in finished drinking waters is going to be with us a long time. It will be with us until we can cultivate the virus of infectious hepatitis and it will be with us until we learn a great deal more than we know now about the slow-growing viroids. I am not aware, however, that these organisms differ structurally from the larger viruses, *i.e.*, they all appear to consist of a nucleic acid in a protein coat. Consequently, we should not expect the viroids to differ very much from the larger viruses in their response to HOCl. However, other factors could contribute to a finding of significant differences, *e.g.*, a small virus could be protected from the action of HOCl by being imbedded in a particulate which would not be large enough to provide the same protection to a larger virus.

Unfortunately, HOCl is a factor in the production of trihalomethanes—substances that we would like to minimize the presence of in our drinking water. Fortunately, we have three other powerful agents that might be considered. Morris (8) reports that ozone is even more effective than HOCl, and that viruses—although more resistant than vegatative bacteria—can also be readily handled by ozone. Chlorine dioxide appears capable of handling viruses, and so does one other technique which is not discussed very often—high pH (12) With all these techniques available, there appears to be ample methodology for the handling of viruses in a reuse situation. The concern really boils down to other qualities of the water and whether these might interfere with the disinfection process.

3) Pseudomonads

Most of us are aware of the ubiquity of the pseudomonads. I think they can be found almost anywhere that you look. Alexander (2) states that only a small number of bacterial species can bring about denitrification, and of these, pseudomonads—including the pathogenic *Pseudomonas aeruginosa*—are the more important. Possibly because of their relationship to denitrification, pseudomonads are important organisms in soils and in wastewater treatment.

Even though the pathogenicity of *Ps. aeruginosa* is determined more by the patient's state of resistance than by any inherent virulence (9), the thought of creating conditions for these microorganisms to proliferate in our distribution and plumbing systems is cause for some soul-searching in any discussions of reuse for potable purposes.

Recommendations

Drug-resistant bacteria

1. EPA should support periodic surveys of the drug-resistance of coliforms isolated from public water supplies. By doing this, a picture should evolve which will better define the potential of the problem to our water supplies.
2. EPA should not adopt new coliform standards, but should take a hard-nosed stance about any infraction of existing standards. The monitoring program for example, might call for the following:
    a) If a coliform determination is >1 or >2.2 per 100 ml (depending upon methodology), immediate resampling should be done. (If it is above 4/100 ml, public notification should be made)
    b) If the resampling is >1 or 2.2 per 100 ml, both public notification and immediate study should be done.

Viruses

1. Any studies of virus presence in finished waters must pay more attention to those confounding factors that are known to interfere with the disinfection processes used. Turbidity, pH, and temperature are not the only factors; NH_3 organic-N, chlorine species, a measure of organics, and particle-size are also important (intuitively).
2. Routine coliphage determinations at select water treatment plants should be instituted. If the disinfection process is working as it should, there should be no viable coliphage in the finished water. If coliphage are found, why look for pathogenic viruses? The conditions for pathogenic virus survival will have existed.
3. EPA should support basic studies of viruses and basic studies of virus disinfection by a variety of disinfecting or inactivating methods.

Pseudomonads

The problem which I have referred to as a *Pseudomonas* problem should really be addressed as a problem of maintaining water quality in distribution systems. Recommendations for EPA activity here would include:

1. Basic studies of water quality and organism growth.
2. Strict application of the rules of sanitary surveys with regard to covered reservoirs, dead ends, adequate pressures, main disinfection, etc.
3. Studies of the biological stability of reclaimed effluents which are, in turn, being studied for potential reuse.
4. Application of the total plate count to the monitoring of select distribution systems. These might be selected on the basis of a sanitary survey, an organic content, and/or on the basis of seasonal effects.

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EVALUATION OF MICROBIOLOGICAL STANDARDS FOR DRINKING WATER

Engineering Control Practices

by

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Introduction

It seems appropriate to hold this symposium at a time when one of the foremost citizen concerns of at least half the Nation is water, or perhaps, more accurately, the lack of it. Regional water crises are not new. In my lifetime, I have acquired some knowledge of several, and have had personal experience with serious water shortage and water quality problems in the West and Midwest. Assuming my remarks are of some interest and significance to symposium participants, please, in assessing them, keep in mind my western and middlewestern perspective (or provincialism, if you will).

My experience has been in the fields of public health—state sanitary engineering and the U.S. Public Health Service—teaching certain sanitary engineering subjects and, in more recent years, the practice of engineering in a professional engineering firm specializing in energy and water projects. I emphasize energy because water and energy are closely intertwined. Each impacts the other.

With this brief background, I shall now proceed to the business at hand, engineering control practices, with the full knowledge that I am tackling a complex matter that cannot be brought into focus in 20 minutes, or even 20 hours or 20 days. Therefore, please bear with me while I report briefly on what is expected of professional engineering and water utility managers and operators in the vital fields of public water supply and water quality control. I shall make occasional reference to Dr. Murphy's law and its corollaries.

Drinking Water Standards—Past, Present, and Future

To navigate successfully, one must have good charts, know where he is going, where he is, and where he has been. There must be a few questions about where we are and where we are going or we wouldn't be holding this symposium. Hopefully, at its conclusion, we shall have some better charts than now. Let's examine briefly where we have been and where we seem to be.

The first U.S. drinking water standards that enjoyed official status were those established by the U.S. Public Health Service in 1914. Often called the "Treasury Standards", they covered only the bacteriological aspects of water quality. A most interesting feature of these standards was the inclusion of a limit for the "standard plate count". Why was it included? Why was it dropped? What is its importance?

The 1914 standards bulletin, which sold for five cents, briefly discussed the sanitary survey and its importance. The sanitary survey, well known to all older sanitary engineers, has been a unifying influence in the standards for over half a century.

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TABLE 1. CHANGING STANDARDS FOR DRINKING WATER.

[Tabulated numerical values are in milligrams per liter (mg/l) except for Bacteria, pH, Taste/Odor, Turbidity, Year, Color]

	1914	1925	1942	1946	1962	1975-1977 *
Coliform Bacteria/100 ml	<2	<1	<1	<1	<1	<1
Total Bacteria/ml	100	—	—	—	—	—
Copper	—	0.2	3.0	3.0	1.0	1.0
Iron	—	0.3	0.3	0.3	0.3	0.3
Lead	—	0.1	0.1	0.1	0.05	0.05
Manganese	—	0.05	0.05	0.05	0.05	0.05
Magnesium	—	100	125	125	—	—
Zinc	—	5.0	15	15.0	5.0	5.0
Arsenic	—	—	0.05	0.05	0.05	0.05
Chromium	—	—	—	0.05	0.05	0.05
Selenium	—	—	0.05	0.05	0.05	0.01
Barium	—	—	—	—	1.0	1.0
Cadmium	—	—	—	—	0.01	0.01
Mercury	—	—	—	—	—	0.002
Silver	—	—	—	—	—	0.05
pH	—	—	<10.6	<10.6	—	6.5–8.5
Chloride	—	250	250	250	250	250
Sulfate	—	250	250	250	250	250
Total Dissolved Solids	—	1000	500	500	500	500
Taste/Odor (Threshold Number)	—	None	None	None	3.	3.
Turbidity (Turbidity Units)	—	"Clear"	10.	10.	5.	1.
Color Units	—	"Colorless"	20.	20.	15.	15.

Hydrogen Sulfide	—	—	—	—	—	0.05
Fuoride	—	—	1.0	1.5	0.8–1.67	1.4–2.4
Phenols	—	—	0.001	0.001	0.001	—
Cyanide	—	—	—	—	.01	—
Nitrate (N)	—	—	—	—	10.	10.
Foaming Agents	—	—	—	—	0.5	0.5
Carbon Chloroform Extract	—	—	—	—	0.2	—
Radioactivity	—	—	—	—	Yes	Yes
Pesticides	—	—	—	—	—	.0002–0.1
Sanitary Survey	Yes	Yes	Yes	Yes	Yes	Yes
Viruses						?
Asbestos						?
Chlorinated Organics						?
What is next?						?

* Federal Register, Dec. 24, 1975; July 9, 1976; March 31, 1977.

Table 1 is an attempt to summarize past Public Health Service (1914-62) and present Environmental Protection Agency standards. This table reflects some sixty years of changing culture. It is apparent that bacteriological quality is still of concern but the bacteriological standards have not changed greatly since 1914. Probably an engineer who could design a plant meeting the 1914 coliform standards could readily design one that could meet current coliform standards although much has been learned about water supply and treatment since 1914. The table also tells us that, over the years, there has been greatly increased concern over things of a chemical, or better, a microchemical nature. This upsurge in interest in microchemistry is essentially a post-World War II happening. This era was marked by an upsurge in industrial chemistry, notably petrochemicals, and wastes associated with their manufacturing and use were bound to have an impact on the water resources of the world. Parenthetically, it should be noted that these new chemical products were welcomed by consumers who found them effective and convenient. Accompanying this industrial revolution was almost overwhelming progress in analytical chemistry, which is truly the cornerstone of knowledge about this complex, universal solvent we call "water". We have, in about 30 years, moved from the era of microbiology into microchemistry. Yet, with the emphasis on chemical quality, we must not forget, not for a moment, the fundamental and continuing importance of microbiological water quality. Speaking very generally, microbiological contamination produces acute health effects. In a susceptible population, water-borne disease will often manifest itself in a matter of days or weeks. On the other hand, usual microchemical pollution; i.e., excluding chemical accidents involving massive contamination, is chronic in nature, sometimes requiring years to make its effects felt. I repeat, these effects, while very serious, are usually long-term rather than short-term. This, of course, makes the investigation of microchemical contaminants exceedingly difficult.

Regarding the future of our drinking water standards, I find the crystal ball quite cloudy. I am certain, however, that they will reflect the inexorable march of science. They will be more complex, not less. The difference between the standards of the year 2000 and those of the present probably will be greater than the difference, noted in the table, between 1914 and 1977. (The proposed upper limit for pH given in the new standards seems unreasonable.)

I would be remiss if I did not remind you that at least one very important facet of the standards does not lend itself well to numerical expression. Consequently, it is not included in the table I have presented. This is the statement, and I quote from the 1962 standards: "The water supply should be obtained from the most desirable source which is feasible, and effort should be made to prevent or control pollution of the source. If the source is not adequately protected by natural means, the supply shall be adequately protected by treatment".

The Microbiology Standard

This symposium concentrates on microbiological water quality, which is quite appropriate. I am sure the organizers recognized the impossibility of discussing microbiology, per se. In thinking about the microbiology of drinking water, it is essential to think of many other matters, not the least of which is disinfection. As is so often the case in this imperfect world, the solution of one problem can create another. The notable example is chlorination for bacteriological quality improvement which can be attended by potentially harmful side reactions. Chlorine has a good bacterial toxicity but a poor chemical specificity, and this fact is very important in many situations.

Over the years the coliform test has served us well and I have little doubt that it will continue to do so. It is by no means perfect, but what is? The standard test which is the heart of the standard itself may be criticized as covering too broad a spectrum, too difficult and time consuming, too sensitive in some respects, not sensitive enough in others. True, the standard test has on occasion caused a false alarm and much embarrassment but this has been the result of trying to short-cut the standard procedure and not that of any inherent defect. Yet, for all of this, the coliform test and the coliform standard have certainly been a major force in our conquest of epidemic disease.

As a state sanitary engineer, I was once responsible for checking the quality of some 400 public water supplies. My predecessors had done a good job and in general these supplies were quite satisfactory from a sanitary standpoint. Most were from protected wells. Water from surface sources, with one exception, was filtered. Water samples collected by health department personnel and those shipped to the department laboratory by water utility personnel were competently examined and the results promptly transmitted to the utility. "Bad" samples, i.e., those showing coliforms, were reported by telephone or telegram and confirmed by letter. Quarterly summaries of the bacteriological quality of all supplies were prepared and posted. It was dismaying to note that each quarter, some 100 of the supplies failed to meet the drinking water standards. What was the reason for this rather alarming state of affairs? I hate to admit it, but I couldn't answer this question then and I can't now. I can only make a few clarifying comments. As I recall, none of the surface water supplies, all subject to microbiological contamination, and treating waters containing from about 10 to far above 100,000 coliform organisms (MPN) per 100 ml, ever failed to meet the standards. Neither did those ground water supplies where fairly extensive treatment systems had been installed; e.g., iron removal, softening. The principal things in common about these utilities were filtration and chlorination. Of these two, I did then and do now, believe chlorination was of major importance. Invariably, the supplies that failed to meet the standards were unchlorinated supplies derived from wells. In general, the wells were properly located and protected (there were, of course, exceptions) and they supplied water that met the bacteriological standards. However, we encouraged water utility personnel to obtain samples from various points throughout the distribution system and they complied. The problem of "bad" samples could, in general, be identified with the following:

1. Poor sampling
    a. bad technique
    b. improper sampling point; e.g., a fire hydrant.
2. Water main, service line, plumbing repairs; failure to flush and disinfect.
3. Tank painting and repairs; failure to flush and disinfect. Improper vents and access covers.
4. Well, well pump, etc. repairs; failure to flush and disinfect.
5. Cross connections; backflow problems.
6. Low or negative pressures in system because of operating defects or inadequate hydraulic capacity.
7. Ground storage repairs; improper vents, covers, access manholes.

Other than bad sampling, which is a matter of training and judgment, the problems seemed to arise principally from minor contamination in the distribution system. Now, there were probably about as many defects in the distribution systems of surface sources as there were in ground sources. Yet the surface supplies had good records while many of the unchlorinated ground supplies had poor records. The difference apparently was chlorination. The results of a campaign to secure chlorination of all supplies supported this. When chlorination was applied, a system that formerly gave an unacceptably high proportion of bad samples suddenly turned around and consistently met the standards. Simple, continuous chlorination of all wells made the difference, which was dramatic. In fact, the clean-up was so sudden and complete that the state sanitary engineer was accused of falsifying bacteriological data simply to prove his point that chlorination would go a long ways toward improving water safety. The conclusion: the bacteriological standards can be met, in fact exceeded, by the exercise of reasonable care in system operation coupled with simple continuous chlorination. To summarize, most of the supplies involved were acceptable until they entered the distribution system. Here, minor contamination occurred from a multiplicity of sources. Chlorination was generally adequate to insure compliance with the standards then in force, which were essentially the same as those published by EPA in 1975. As a brief epilogue, I can report that the state's program of insistence on chlorination of all supplies was continued and as of today, all public water supplies, over 700, are chlorinated.

As to surface waters, the program included "selling" free-residual prechlorination to surface water utilities. Generally, this was well received. Chlorine should be recognized as a water treatment chemical. It is more than a disinfectant. Prechlorination to a free residual provides values beyond those associated with excellence in disinfection.

I do not expect to pursue the matter of nematodes in drinking water but they have been found and they may be more common than we realize in supplies derived from some surface waters. Their source probably is the raw water, but the possibility that they may find a "home" in the treatment plant cannot be excluded. Nematodes are macroscopic, easily detected, and resistant to chlorine. Their greatest health significance seems to be the fact that they may ingest pathogenic bacteria and then serve as a carrier and protector for these pathogens. The water industry is very much aware of nematodes. They are indeed a "can of worms". Chang and others have conducted extensive investigations of nematodes in water.

Coliform criteria have been proposed by organizations other than EPA. The American Water Works Association has established "quality goals" for drinking water and the AWWA rationale states: "Modern disinfection control procedures are such that a practical goal can be destruction of all coliform organisms". The quality goal is "no coliform organisms". This is, I think, an admirable goal, but proof of compliance probably would be virtually impossible. We cannot deal in absolutes in the water safety business.

The City of Chicago, well known for excellence in the control of water treatment systems, adopted a "Chicago Standard" of not more than 0.5% of 10 ml standard portions positive in contrast to an allowable 10%.

There is some relationship between raw water quality and that of the finished water. Over the years, guidelines for maximum allowable raw-water coliform concentrations have been recommended. These have posed problems and, in large measure, have been ignored, especially on "flashy" midwestern streams where coliform levels are usually fairly low during low and moderate flow periods but go out of sight during high flow periods. The latest coliform criterion for raw waters is that of the National Academy of Science—National Academy of Engineering published in the "blue book" in 1973. This calls for a coliform limit of 20,000/100 ml (2,000/100 ml fecal coliform). It should be understood that this limit is based upon a "defined treatment process", as specified in the NAS-NAE publication "Water Quality Criteria 1972". With additional treatment, a much higher coliform level can be handled without difficulty.

We need to understand that "coliforms" represent a fairly broad spectrum of organisms derived from many sources, not necessarily disease related. Studies (1970) by the Department of Health and Environment, State of Kansas, (Soldier Creek Basin), have shown that the "heaviest bacterial pollution (coliforms) existed during high flow periods" when dilution was the greatest. Natural and agricultural sources were obviously significant contributors. The watershed had an area of 290 square miles and consisted principally of some 400 farms plus a 120 square mile Indian Reservation, largely undeveloped. There were no "point sources" of pollution in the watershed; i.e., no municipal or industrial outfalls and no confined animal feeding operations.

I am especially familiar with water treatment plants located along the Missouri River between Omaha, Nebraska and the Missouri-Mississippi confluence at St. Louis. These plants at times "see" very high apparent coliform densities yet have no difficulty meeting the bacterial standards. They are, of course, operated by competent personnel and provide multistage treatment including pre and post chlorination. Many of the plants also include lime softening, a process long known for its effectiveness in the reduction of bacterial densities.

I believe the coliform test has been and still is a valuable water quality monitoring tool. It has some shortcomings which are well known to this audience. Despite these, if I were faced with a decision regarding water quality and the constraint of being allowed only one test, I believe the coliform test is the one I would select. In applying the test to treated water, one short-cut to sure trouble is failure to carry the test through the "completed" phase when coliforms are "confirmed" in a finished water in which they are normally not found. One of my gripes about the standard test is the "confirmed" designation for the second phase. The word "confirmed" is a strong word and easily misinterpreted. Coliforms are not "confirmed" until the "completed" test is conducted—a rather complex and time-consuming procedure. I wish we could come up with a truly descriptive word for the "confirmed" test.

The relationship between coliform density and pathogens is understood principally in a qualitative sense. Some thirty years ago a paper was published in which the authors, Kehr & Butterfield, considered the relationship between coliforms and enteric pathogens. For a typhoid rate of .01 to 30/1000 population/year, Kehr & Butterfield suggested a *Salmonella typhi (E. typhosa)* per million coliforms ratio of 3 to 120. They presented evidence that this ratio might remain relatively constant through natural purification processes, approximating 99.9%. They concluded that their studies ". . . emphasize the basic value of the coliform test as an indicator of the possible presence of pathogens and indicate that a very real danger may exist when coliforms, in even moderate concentrations, are present. The factor of safety provided by the ratio of a million or so coliforms present for each *E. typhosa* would, it is believed, take care of the usual fluctuations in the ratio of *E. typhosa* to coliforms provided the density of coliforms . . . be kept quite low or eliminated by methods which reduce the general bacterial population". I commend this paper to you for its interest and the imagination displayed by the authors, both of whom I knew and liked as individuals and respected as competent scientists.

In regard to viruses, Berger, et al, reporting for the American Society of Civil Engineers in 1970, summarized water treatment practice as follows:

> "Ideally, the water treatment plant should be capable of reducing turbidity to less than 0.1 Jackson Unit and provide an HOCl residual of 1 ppm after contact period of 30 minutes. In practice, it may be assumed that for waters having a pH less than 8, an acceptable level of protection will be provided by treatment producing a turbidity less than 5 Jackson Units and a free chlorine residual of 0.3-0.4 ppm with a contact period of 30 minutes. For waters with higher pH, either the chlorine residual or the contact period should be increased by half. Because the water treatment plant is the major line of defense against introduction of viruses into the drinking water system, it is necessary to emphasize the absolute necessity of constant vigilance in careful plant operation."

I believe these recommendations still provide sound guidance to the engineering profession.

My conclusions: Chlorine is a proven disinfectant. Prechlorination to a free residual provides excellence in disinfection and is a valuable treatment adjunct. Ground water supplies need at least marginal chlorination as a safety factor. Such supplies, if not chlorinated, will have difficulty complying with existing bacterial standards, principally because of "problems" associated with distribution. As of now and assuming we can continue to use chlorine as in the past, I see no serious problems in complying with the intent of the standards.

Supplementing these remarks, I do foresee some problems for the small utilities who collect only one sample per month. Chance contamination resulting from faulty collection, poor judgment in selecting the sampling point, and laboratory errors can easily throw a small utility out of compliance. This sets up a chain reaction of notification that could prove extremely burdensome and possibly result in loss of public confidence if repeated frequently. Public notification has both good and bad features. It is a delicate matter. Judgment is involved in this entire matter of standards compliance and we all know the futility of trying to write judgment into any sort of law, rule, regulation, or guideline. Some better publications about the standards would be helpful. I commend Region VII, EPA, for their booklet titled "Answers to Questions About the Safe Drinking Water Act". Hopefully, the standards themselves will be pubished in better format than that provided by the Federal Register, which is poor.

Water Distribution

Here is a real problem. The distribution system, the most costly portion of a public water supply, has in full measure been treated like a "poor relative" in the past. This attitude is now changing. There can be many troubles in the distribution system. I have already mentioned some of them. Dr. Murphy's law, which tells us that: "In any field of scientific endeavor, that which can go wrong will go wrong" certainly finds application in the water distribution system.

The distribution system presents a large water-pipe interface. I once roughly estimated this at 0.6 to 0.7 acre per 1,000 population. Strange things can and do happen at this very large interface. Corrosion is one, but in keeping with the theme of this conference, I shall emphasize biological effects. These effects, which produce dramatic water quality changes, are identified in the literature and reach their peak in distribution zones of poor circulation, notably "dead ends". Here chlorine disappears and remaining bacteria make the most of their opportunity to grow and produce changes, all of which are displeasing to the water customer. Perhaps there is no health problem but the customers don't like turbid, smelly water and they won't suffer it in silence either. On the other hand, perhaps there are health problems. These bacteria are seldom studied to any extent. Possibly they have greater direct or indirect health significance than we realize. An indirect effect that has been identified is interference with the standard coliform test. This reminds me that the present and past standards, the 1914 edition excepted, are silent on the matter of the total numbers of bacteria allowable. I know there are good reasons for this omission; but, nevertheless, consider it unfortunate. As an alternative, I hope EPA will strengthen its statements and recommendaions as to the value of the "standard plate count" and will also recommend a guide number. If I were running a water system, I would certainly include the plate count as a routine test (9).

Another problem in water distribution, the significance of which has not been generally appreciated, is that of bacterial slime formation on the interior of water transmission mains. This is serious for at least two reasons: the hydraulic capacity of the pipe is significantly reduced; energy requirements for pumping are significantly increased. It is almost unbelievable what a thin, rippling deposit of bacterial slime will do to a pipe's flow characteristics. A very thin, rippling, slime layer averaging no more than about 1/32 inch in thickness can reduce the capacity of a fairly large pipe as much as 50% without appreciably changing its internal diameter. The slime growth is microbial in nature and has high adsorptive capacity. It often contains considerable iron and manganese. The slime forming organisms can live and thrive on trace amounts of organics and minerals. The film builds insidiously over a period of years by extracting nutrients from the water and excreting waste products into it.

Consider a 12-inch water pipe, connected to a pumping station. The pipe, when installed, has a Hazen-Williams "C" value of 135. If, in five years, this "C" value is reduced to, say, 80, the capacity *loss* for the same pressure drop is $100(135-80)/135 = 41\%$. To maintain the original flow in the pipe, the pressure at the pumping station must be increased to the extent of $[135/80]^{1.85} = 2.6$ times its initial value or an *increase* of 160%. Considering that energy and energy costs, associated with pumping, vary directly as the pressure against which the pumps are working, it is evident that bacterial slime growths in pipelines are an extremely serious matter. These slime, or so-called "nuisance", organisms may or may not have health significance. (I doubt that we can answer this question entirely satisfactorily.) On the other hand, they do have practical operating and economic significance. The alternative to higher pumping pressures is to clean the line and restore the "C" value. This is expensive and difficult, and, obviously, the pipe must be out of service during cleaning. Furthermore, cleaning is, at best, a temporary remedy. Unless the organisms are controlled somehow, the whole process starts over the day the clean pipe is put back into service. Pipe slime brings the water-energy relationship into sharp focus.

Another objectionable occurrence that can result from pipe slime is a sloughing off of quantities of the slime. This can produce an offensive and probably unwholesome condition at what the U.S. Public Health Service, and now the EPA, call the "free flowing outlet of the consumer".

The distribution system is indeed a labyrinth, replete with traps for the unwary. It often illustrates a well known corollary of Dr. Murphy's law: "Left to themselves, things always go from bad to worse".

The Anatomy of a Water-Borne Epidemic

Let's look at what can happen in distribution. This "happening", or one like it, might well form the topic of another symposium. My treatment of it will be necessarily brief. This city, located in the western Midwest, had a population of about 11,000 and was an important railroad center. (The latter was important in at least two respects.) The water supply was obtained from eight wells having a depth of somewhat more than 100 feet. They were located several miles from the city and tapped a major aquifer known to be of good mineral and biological quality. The well water was pumped to a concrete storage reservoir, covered, as I recall, from which it was repumped through about 8 miles of 12 inch and 14 inch transmission pipes to the city's water distribution system. Early in September, the state health department was notified by a practicing physician that a severe outbreak of intestinal disorder was occurring in the city. Samples of water were collected and coliform tests performed. Samples from the wells were "clear". Samples from the distribution system all contained fairly large numbers of coliform bacteria. The water system did not include chlorination, and emergency chlorination equipment was installed. "Boil-the-water" advice was distributed. The railroad, a major line, was ordered to cease taking on city water. World War II was in progress and war industries and members of the armed forces traveling on troop trains were involved, as well as the city population. Corrective efforts were too late to avert an epidemic and there were more than 3,000

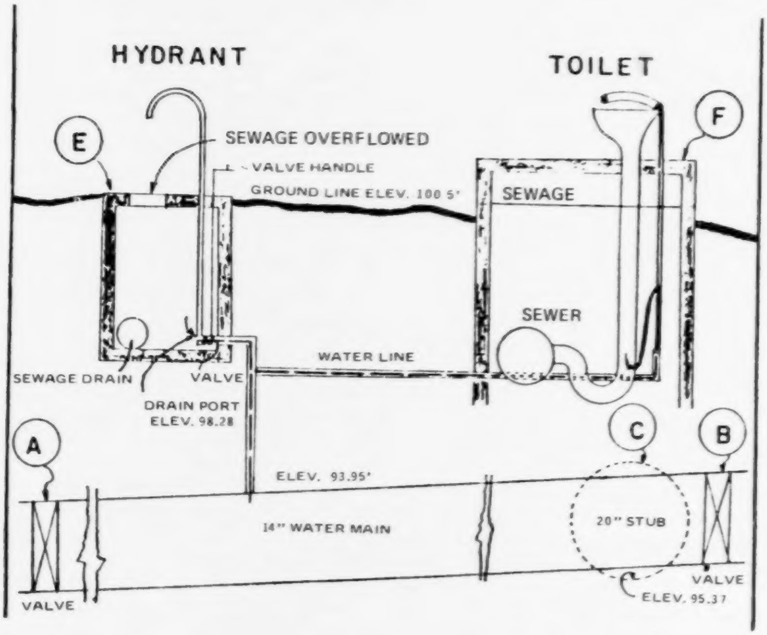
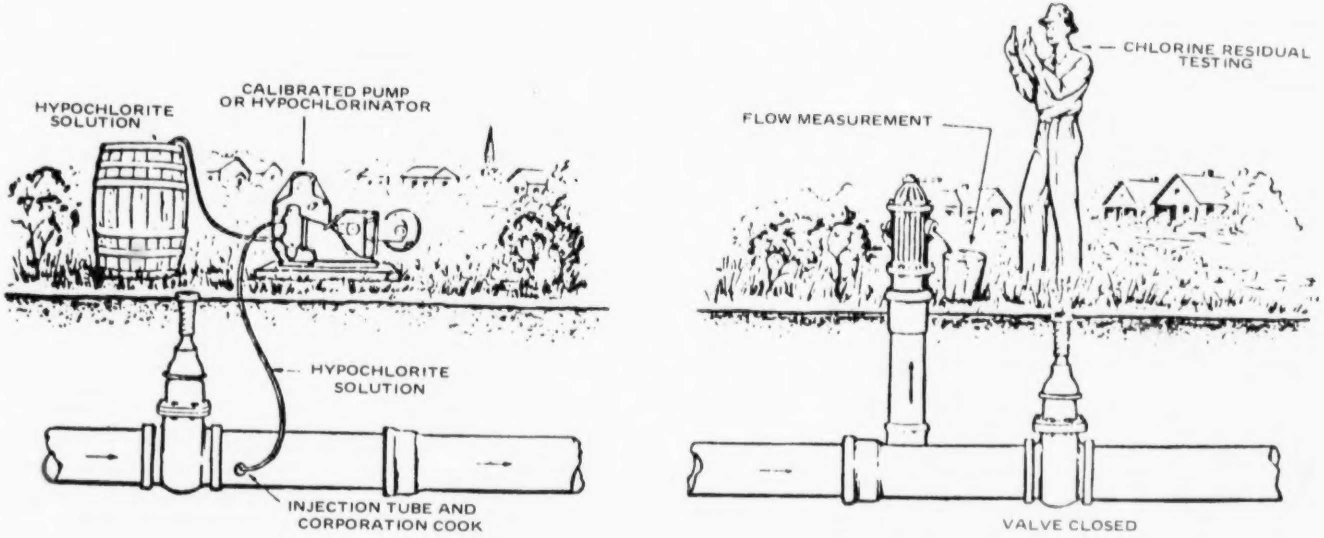

HYDRANT
TOILET
E
F
SEWAGE OVERFLOWED
VALVE HANDLE
GROUND LINE ELEV. 100 5'
SEWAGE
SEWER
WATER LINE
SEWAGE DRAIN
VALVE
DRAIN PORT
ELEV. 98.28
A
C
B
ELEV. 93.95'
14" WATER MAIN
20" STUB
VALVE
VALVE
ELEV. 95.37

Figure 1. CROSS - CONNECTION SCHEMATIC DRAWING.
(American Journal of Public Health, 1944)


HYPOCHLORITE SOLUTION
CALIBRATED PUMP OR HYPOCHLORINATOR
HYPOCHLORITE SOLUTION
INJECTION TUBE AND CORPORATION COOK
FLOW MEASUREMENT
CHLORINE RESIDUAL TESTING
VALVE CLOSED

TYPICAL SET UP FOR WATER DISINFECTION WITH A HYPOCHLORINATOR.

Figure 2. DISINFECTION OF NEW OR REPAIRED WATER MAINS. (Office of Civilian Defense, 1943)

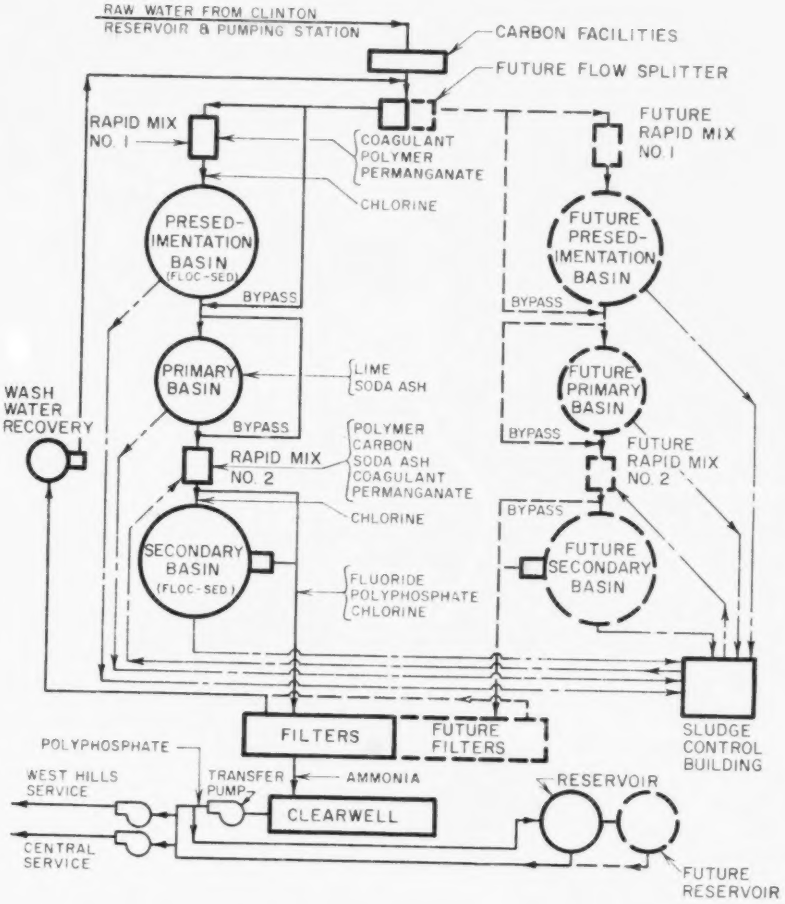

RAW WATER FROM CLINTON
RESERVOIR & PUMPING STATION
CARBON FACILITIES
FUTURE FLOW SPLITTER
RAPID MIX
NO. 1
COAGULANT
POLYMER
PERMANGANATE
CHLORINE
FUTURE
RAPID MIX
NO. 1
PRESED-
IMENTATION
BASIN
(FLOC-SED)
BYPASS
FUTURE
PRESED-
IMENTATION
BASIN
BYPASS
PRIMARY
BASIN
LIME
SODA ASH
FUTURE
PRIMARY
BASIN
WASH
WATER
RECOVERY
BYPASS
BYPASS
RAPID MIX
NO. 2
POLYMER
CARBON
SODA ASH
COAGULANT
PERMANGANATE
FUTURE
RAPID MIX
NO. 2
CHLORINE
BYPASS
SECONDARY
BASIN
(FLOC-SED)
FLUORIDE
POLYPHOSPHATE
CHLORINE
FUTURE
SECONDARY
BASIN
FILTERS
FUTURE
FILTERS
SLUDGE
CONTROL
BUILDING
POLYPHOSPHATE
WEST HILLS
SERVICE
TRANSFER
PUMP
AMMONIA
CLEARWELL
RESERVOIR
CENTRAL
SERVICE
FUTURE
RESERVOIR

Figure 3. LAWRENCE, KANSAS - CLINTON WATER TREATMENT PLANT FLOW DIAGRAM AND POINTS OF CHEMICAL APPLICATION.

cases of dysentery. Public health laboratory tests showed that the predominant organism was *Shigella paradysenteriae*. How did this organism enter the system? Figure 1 gives essential details. A 14 inch supply main was out of service for repairs. Pressure was "off" for 12 hours or more. During this period there was a sewer stoppage in a small railroad village where railroad maintenance employees were quartered. The sewage backed up, overflowed frost-proof hydrant pits and flowed backward into the 14 inch transmission main. The head available to cause this flow of sewage into the empty transmission main was at least six feet. How much sewage entered the water main is unknown, but it was enough. When the transmission main was restored to service, it was given only a minimum flush-out and was not disinfected. Recalling that this was a transmission main, when service was restored, whatever sewage was stored in the main, probably quite a bit, was distributed throughout the city. Here is a thing that should not have happened and the probability of its occurrence was very low. Nevertheless, it did happen. Here is a distribution system disaster that chlorination of the supply would not have averted. One lesson derived from this experience is that pipes out of service for repair must be flushed thoroughly and disinfected before they can be assumed to furnish safe water. How often is this procedure followed? Often? Seldom? Not at all? The answer is, I believe, "often" but not "always". Remember, a water main out of service is a serious matter and there is great pressure to restore service for public fire safety as well as convenience. It is the duty of responsible professionals to guide repair crews as to the correct procedure. Disinfection of a new or repaired main is no mean feat. It often requires mechanical ingenuity and technical competence of a high order (Figure 2). Another lesson in "preventive medicine" is that if "frost-proof" facilities are installed, they should not be drained to a sewer.

Referring again to Murphy's Law, this disaster seems to be an illustration of the important corollary: "If there is a possibility of things going wrong, the one that will go wrong is the one that will do the most damage". The distribution system often seems to be a living, thinking thing, well aware of all of Murphy's laws and how to apply them.

If you are not familiar with water works practice and want to know more about it, I recommend to you an old publication, produced by the Office of Civilian Defense during World War II (1943)—OCD Publication 2022, titled "Waterworks Engineering in Disaster".

The Lawrence, Kansas Clinton Reservoir Water Treatment Plant

Figure 3 illustrates this water treatment plant, recently designed by the author's firm. It will have a capacity of 10 million gallons daily and includes softening, as well as other standard treatment systems.

The raw water will be obtained from Clinton Reservoir, a 7,000 acre lake, now being constructed by the Corps of Engineers, four miles southwest of Lawrence, Kansas. This is a multi-purpose reservoir. Its functions include: flood control, water quality control, recreation, public water supply, and biosupport. The dam will control 367 square miles of the watershed of the Wakarusa River. Storage allocations are as follows:

Flood Control	267,800 acre-feet
Multipurpose	129,400
Sediment	28,500
Water Supply	110,400 (36 billion gallons)

Land use in the watershed is principally crop land and pasture. Two small, sewered communities are located about 15 miles above the reservoir. Their combined population is about 2500. Both have sewage treatment plants. There are also a few rural mobile home parks. There are no significant agricultural or industrial waste point sources. Estimated mineral characteristics of the impounded water are as follows:

Long-Term Mean Values

Cations			*Anions*		
	mg/l	meq/l		mg/l	meq/l
Ca^{2+}	88	4.39	HCO_3^-	280	4.59
Mg^{2+}	14	1.15	SO_4^{2-}	69	1.44
Na^+	14	0.61			
K^+	3.4	0.09	Cl^-	10	0.28
		6.24			6.31

Total hardness is expected to range from 100 to 382 mg/l, carbonate hardness, 100 to 332 mg/l. Coliform and fecal coliform densities will vary widely with runoff. Coliform MPN values in the range of 200,000 to 300,000/100 ml are likely during peak inflow periods. The estimated long-term fecal-coliform, goemetric-mean density is expected to be about 50/100 ml. Probably the most difficult water treatment conditions will be those associated with algal blooms and related taste-and-odor, coagulation and filtration problems. Taste-and-odor control will, as usual, be the most difficult and the most unpredictable.

Note from the schematic that free residual chlorination will be employed, with provision for alternative chlorine feed points. The water will be ammoniated after filtration and the free chlorine converted to chloramine.

Detention time in the various basins at 10 mgd will be as follows:

	Hours
Presedimentation	3.1
Primary *	2.4
Secondary	3.1
	8.6

* Based on 20% "split" treatment bypass

This is a symposium on microbiology and is, therefore, concerned with all those things that contribute to rendering the water "safe". I have no feeling of uncertainty about the bacterial quality of the water this plant will deliver to public service. We should have no difficulty meeting the coliform "MCLs". In fact, I would be surprised if we did not do much better than the drinking water standards require. For viruses, we have no standards, but I believe that if we did have, the plant effluent would readily meet them. The plant will employ excess lime treatment and high lime treatment has been shown to be quite effective in virus reduction. This may be due to the combination of high pH plus the excellent coagulation achieved by lime softening of a fairly hard water. In addition, we have free-residual chlorination and considerable contact time. We also have, as back-up, dual media filters. If there are any viruses present, we should remove them, destroy them, or at least inactivate them to the point that they are incapable of producing disease. While I am no authority on viruses, my judgment, based on experience with water plants and literature reports, tells me this plant will produce "safe" water. The water should also be esthetically pleasing and soft enough for general domestic use. The turbidity standard also can be met without difficulty. Continuous turbidity monitoring equipment has been provided.

Impact of the Standards on Engineering Practice

Turbidity, a light scattering phenomenon, is closely related to microbiological quality. When turbidity is measured, we are indirectly measuring particulates in the water and particulates are important. Turbidity has long been viewed as a sign of potential danger. Certainly, turbid water, whatever the cause, is unappealing, if not actually repulsive. Good turbidity removal at a treatment plant is reassuring insofar as bacterial and virus hazards are concerned. Turbidity is appropriately classed as a health hazard and while not all turbidity is related to microbiological quality, often it is closely related. Turbidity in finished water may interfere with chlorine disinfection, prevent maintenance of a residual during distribution, interfere with bacteriological testing, and be a sign of haphazard plant operation and consequent danger of water-borne disease.

I do not believe there is anything in the current EPA microbiological drinking water standards that changed the design of the Lawrence, Kansas plant. If, in the future, a virus standard is set, monitoring for viruses will be required, and this will call for additional effort by laboratory personnel. There is, however, looming on the horizon the matter of microchemical effects of chlorination. Organic chemists have long known of chlorine's affinity for organics and its ability to enter organic molecules, forming a variety of chloro-substitution products. Only recently have health authorities and the water industry begun to delve into these rather subtle, attenuated chlorine-organic reactions in water. We had some experience in the past with chlorophenols but this was considered principally a taste-odor problem without health significance. Regrettably, we didn't look farther, but the analytical equipment and the competence to run it have not been generally available for very many years.

We are now confronted with the real possibility of a standard for halogenated compounds in water, such as: carbon tetrachloride, chloroform, dichloromethane, bromoform, dibromomethane, and probably others. This standard doubtlessly will set limits in terms of micrograms rather than milligrams per liter, or possibly micromoles per liter. If this standard cannot be met by conventional chlorination practice, then modifications will be required; however, in making these modifications, we must be ever mindful of their effects on microbiological quality of the finished water. The new microchemical standards involving halogenated organics are closely related to the microbiological standards because they may influence, and possibly drastically alter, chlorination practice. Engineers and the water industry have had a lot of experience with chlorine and we are not going to cast it aside without a great deal of careful consideration. It is in this area that engineering practice and drinking water standards are likely to collide head-on. Remember, I am speculating about a probable standard of unknown magnitude, not an existing standard. If we do, in fact, have to curtail chlorination somehow, or use untried substitutes, there will be great professional concern about microbiological quality. None of us want to solve a microchemical water problem and at the same time create a microbiological hazard. Surely, this would be the negation of sound professional practice.

What can be done? Are there chlorine substitutes that avoid the microchemical problem? There apparently are and they are being studied. Examples are ozone and chlorine dioxide. Both are powerful disinfectants that insofar as we know avoid the possibly harmful organic reactions that result when we use chlorine. Both of these substitutes have engineering and possibly other disadvantages. First, although, we have had some experience with chlorine dioxide, we have not had much, and even less with ozone. The latter has been used for many years in Europe and elsewhere, but there has been little real engineering experience with its generation and application in U.S. water works practice. Other potential problems with ozone are lack of a residual and the inability to store reserve supplies. Ozone must be generated at the rate used. Furthermore, we know very little about the reactions of either ozone or chlorine dioxide with trace substances, notably organics, in water. They are both powerful oxidizing agents, probably fully capable of reacting with many trace organics. What are the reaction products? Are they innocuous or harmful? I don't believe we can answer this question today, and the answers may be several years away.

What other means are available for dealing with the problems that arise from present chlorination practice? Remove the organics. This can be done by granular carbon filtration but only at an almost staggering cost and only after application of considerable engineering effort. While we know a good deal about carbon filtration, there is probably as much to be learned about practical, day-to-day, reliable operation, as has been learned.

We can reduce the reactivity of the chlorine added to water by converting it, in the water, to chloramine. Here, again, we encounter a penalty. Reduced reactivity means reduced disinfecting power. This means higher residuals and a requirement for much longer reaction time. The problem of higher residuals would be easy to handle, but providing the necessary long reaction time can be very difficult. Also, chloramines are not spectacularly effective for destruction or inactivation of viruses.

Summary

The current EPA microbiological standards are essentially those of past years. They are proven standards that can readily be met by sound engineering design and good operating practice. The standards call for considerable paperwork and record keeping, to which many small utilities are unaccustomed. The required paperwork may be a nuisance but it is not all bad. It is dismaying and alarming to learn by experience how many small water utilities, and some not so small, keep poor or practically no water use, quality, and equipment records and have no up-to-date maps of their water systems. The matter of public notification has good and bad features and, doubtlessly, there will be considerable controversy about this requirement.

Present bacteriological standards have not had a major impact on engineering design. With the aid of coagulation, sedimentation, filtration, and chlorination, we can meet or exceed these standards without great difficulty. We can probably also meet any reasonable virus standards proposed. Achieving the turbidity standard should not be difficult, except for some utilities using unfiltered surface supplies. For those, the technology, and engineering expertise to apply it, are available. The problem is, therefore, one of management, finance, and public support. The job can be done, but consumers must expect to pay more for water than in the past. One of the problems in the water industry is that water, a truly precious substance, has always, in the past, been plentiful and cheap—probably too cheap. This is changing.

Any alteration required in chlorination practice, as, for example, curtailment or elimination of prechlorination, will have a major impact on engineering design. We are ready, willing, and able to apply proven new systems, as required, but we can't do the whole job alone. I am pleased that the scientific community is, at long last, showing much more interest in water. Engineers must look to the scientists to define basic water quality needs and we invite them to work with us in charting a true course. Scientists need the help of engineers and engineers certainly need the help of scientists. Working alone, neither group can be completely effective, but working together, there is very little we can't do.

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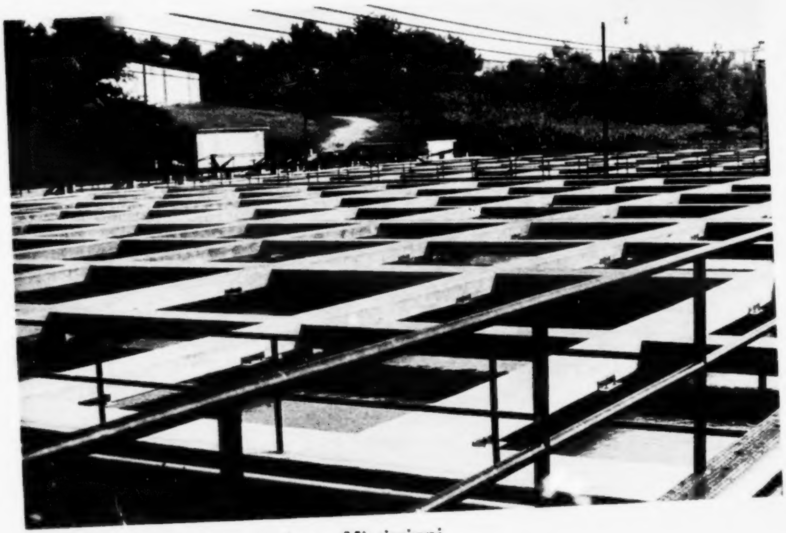

Water Treatment Plant, Jackson, Mississippi

SESSION 3

Compliance with the Coliform Standard

Chairman: Dr. Charles W. Hendricks, Criteria & Standards Division, Office of Drinking Water, U.S. Environmental Protection Agency, Washington, D.C.

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Compliance with Drinking Water Regulations

Charles W. Hendricks
Office of Drinking Water
U.S. Environmental Protection Agency
Washington, D.C. 20460

In 1970, the Department of Health, Education and Welfare undertook a study of public water supply systems (4). The results of this study indicated:

1. 36% of the tap samples contained one or more bacteriological or chemical constituents exceeding the limits in the 1962 Public Health Service Drinking Water Standards.
2. 85% of the systems did not analyze a sufficient number of bacteriological samples.
3. 77% of the plant operators were inadequately trained in fundamental water microbiology.
4. 46% of the plant operators were deficient in knowledge of the chemistry relating to their plant operation.
5. 79% of the systems were not inspected by State or county authorities in the year prior to the study.

For these and other compelling reasons, Congress enacted the Safe Drinking Water Act (2) (PL 93-523) in 1974, "to assure that the water supplied to the public is safe to drink." The basic structure of the Act, as it pertains to public water systems, calls for the establishment of: maximum contaminant levels for most contaminants in drinking water; and criteria and procedures, including quality control measures, to assure compliance with such maximum contaminant levels.

Provisions of the Safe Drinking Water Act and The Interim Primary Drinking Water Regulations

The Safe Drinking Water Act applies to each "Public Water System," defined in Section 1401(4) of the Act as "a system for the provision to the public of piped water for human consumption, if such system has at least fifteen service connections or regularly serves at least twenty-five individuals." Both privately and publicly owned water systems are covered, and the U.S. Environmental Protection Agency has interpreted service "to the public" to include factories, churches, schools, and private housing developments (3).

The definition of a "public water system" in the National Interim Primary Regulations (3) seeks to explain the meaning of the statutory reference to "regular" service. It was originally proposed to interpret the term to include service for three months out of the year, but this definition would have excluded many public accommodations such as campgrounds, lodges and seasonal recreation areas which serve large numbers of tourists but are in operation for perhaps less than three months each year. The definition in the final version covers systems serving at least twenty-five individuals at least 60 days out of the year.

Two types of "public water systems" are recognized in the Interim Primary Regulations—those serving community residents and those serving transients for intermittent users. This differentiation is based on whether the water served is an individual's major source of drinking water. Although this concept has more applicability for chemical than microbiological contaminants, it is, never the less, based upon the fact that the possible health effects of a contaminant in drinking water in many cases are quite different

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for a person drinking the water for a long period of time than for a person drinking the water only briefly or intermittently. For microbiological purposes, however, the risk of infectious disease is dependent on whether the organism is present in the water, and little distinction is made in the Interim Primary Regulations for the microbiological monitoring of very small community water systems serving 1000 or less residents as compared to the non-community supplies which serve the more transient users.

The Maximum Contaminant Levels (MCL), for coliform bacteria contained in the Interim Primary Regulations (Table 1) are based upon the analytical technique used. A laboratory may employ the membrane filter procedure or either the ten or one-hundred milliliter fermentation tube procedure. Any of these procedures may be employed at the discretion of the State, but only one procedure can be used during any compliance period since the limits prescribed for the fermentation tube methods are based upon tubes containing gas and not values taken from the "most probable number" table.

Limits for coliform bacteria, whether the sample has been analyzed by the membrane filter or the fermentation tube technique, are expressed in two parts. The first part of the MCL establishes the arithmetic mean concentration of coliform bacteria that can be allowed in water from the system per month. In essence this number represents a limit for the general water quality of the system as a whole. The second portion of the MCL seeks to place a

TABLE 1. MAXIMUM MICROBIOLOGICAL CONTAMINANT LEVELS *

MEMBRANE FILTER TECHNIQUE

A. A mean of 1 coliform per 100 ml of water per month.
B. Four coliforms per 100 ml of water in more than one sample when less than 20 samples are examined per month; or
Four coliforms per 100 ml of water in more than five percent of the samples when 20 or more samples are examined per month.

FERMENTATION TUBE TECHNIQUE

10 ML Procedure
A. No coliform bacteria in more than 10 percent of the portions examined per month.
B. No coliform bacteria in three or more portions in more than one sample when less than 20 samples are examined per month; or
No coliform bacteria in three or more portions in more than five percent of the samples when 20 or more samples are examined per month.

100 ML Procedure

A. No coliform bacteria in 60 percent of the portions examined per month.
B. No coliform bacteria in five portions in more than one sample when less than five samples are examined per month; or
No coliform in five portions in more than 20 percent of the samples when five or more samples are examined per month.

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* National Interim Primary Drinking Water Regulations (40 CFR 141.14)

limit on the number of coliform bacteria that would be acceptable in any individual sample and represents a limit for water quality at a specific sampling point within the distribution system. Failure of any portion of the limit is sufficient cause to fail the MCL.

Table 2 is a summary of the major sampling and analytical requirements for coliform bacteria that are contained in the Interim Primary Regulations. These provisions instruct the utility in how many samples to take, the analytical procedure to use and under what conditions should follow-up (check samples) be taken. Also included is a provision for the substitution of chlorine residual determinations for some of the required coliform determinations should the States desire to employ it.

The sampling and analytical requirements of the Regulations determine the extent to which compliance with the maximum contaminant levels will be maintained. It has been and continues to be the Office of Water Supply policy to keep the Primary Regulations flexible and to allow for State discretion where possible. For this reason the analytical procedures other than those cited in *Standard Methods* may be employed if it can be shown that the alternative analytical technique is substantially equivalent to the prescribed test in precision and accuracy as it relates to the determination of compliance with the MCL. Likewise, some monitoring requirements may be reduced by the State for small community and non-community systems if local conditions warrant the reduction.

Problem Areas in the Interim Primary Regulations

The Maximum Contaminant Levels for coliform bacteria and their sampling and analytical provisions as contained in the Interim Primary Regula-

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TABLE 2. MICROBIOLOGICAL CONTAMINANT SAMPLING AND ANALYTICAL REQUIREMENTS.*

MAJOR PROVISIONS

A. Analytical procedures are from *Standard Methods,* APHA, 13th Edition.
B. Sampling frequency is based on population and is specified.
C. Sampling frequency for small community systems serving persons 25-1000 may be reduced to 1 per quarter at State discretion.
D. Sampling frequency for non-community systems of 1 per quarter may be reduced on the basis of a sanitary survey.
E. Two consecutive, daily check samples are to be taken if:
  —The MF count exceeds four coliforms per 100 ml of water.
  —Coliforms occur in three or more 10 ml portions.
  —Coliforms occur in five 100 ml portions.
F. Chlorine residual analyses may be substituted for up to 75 percent of the coliform determinations at a rate of four chlorine residual determinations taken for each coliform test substituted. At least one chlorine residual test must be taken per day.
G. Public notification is required if a MCL or a monitoring frequency is violated.

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* National Interim Primary Drinking Water Regulations (40 CFR 141.21)

tions are certainly not the last word in developing drinking water regulations. For example, the promulgated MCL's for coliform bacteria are basically the 1962 Public Health Service Standards (5) with minor refinements and clarifications. However, further changes may be desirable before Revised Regulations are developed. For example, the MCL's for the membrane filter analytical methods do not resolve the question of how many coliform bacteria are assumed to be present in a single highly contaminated sample. Some laboratories assume an upper limit of 50, while others seek to continue to count individual bacteria to a level of 100 or even higher in a single sample. The upper limit assumed will affect the monthly average which is calculated to determine compliance with the MCL's. Another question relating to the coliform bacteria MCL's is the matter of possible spurious positive samples. As the regulations are written, all routine samples must be counted even though subsequent check samples may not indicate contamination. The reason for this is that bacterial contamination is often intermittent or transient. Negative follow-up samples taken a day or more afer a positive routine sample has been analyzed cannot demonstrate that the positive result was in error. It may be possible, however, to prescribe a means of dealing with spurious positive results without compromising the integrity of MCL's.

A third question concerning the MCl's for coliform bacteria is the relationship of monthly averages of coliform bacteria levels to monthly percentages of positive samples. For example, the monthly average MCL for the membrane filter method is violated if the monthly average exceeds one coliform bacterium per sample. However, for purposes of determining whether the month-percentage-of-positive-samples MCL is violated, a sample is counted as positive only if it contains more than four coliform bacteria. Thus, it is possible, particularly when a relatively small number of samples is taken, for a system to fail the monthly average MCL even when no single sample taken during the month is out of compliance with the limit.

In addition to the questions listed above, this Office has been asked to consider an amendment to the Interim Primary Regulations that would specifically address the case where one positive sample would cause the supply to violate the MCL. These requests cite the following.

1. For small utilities (perhaps those less than 10,000 persons served although the cited examples are often those serving 2,400 or less), the collection of one sample requiring the collection of check samples may cause a supply to fail to meet the MCL for that compliance period, thus triggering the public notification requirements.
2. The collection of one such sample followed by two negative check samples should not be counted a failure of the MCL because it could have resulted from improper sampling technique, from accidental contamination in the laboratory, or the lack of precision in the culturing procedures.
3. Public notification by way of the news media would serve no purpose because it would be too long after the fact for consumers to take self-protective actions (even if such action had been justified at the time of collection of the sample in question). The notice may also unnecessarily upset and alarm consumers, and would ultimately be self-defeating as consumers might develop a "ho-hum" attitude when it became apparent no waterborne disease transmission accompanied the contamination notices.

Certainly these comments are significant because they point to an area that is not addressed in the Interim Primary Regulations. I would hope that discussion at this symposium would focus on what to do about false positive results especially for small systems taking less than ten samples per month. In addition to this question, the following also warrant discussion and comment.

1. Validity of the Coliform Indicator System
   —Are we using the proper indicator system?
   —Do the present limits protect health?
   —Is it possible to calculate risk from these limits?
   —Can non-biological tests be substituted?
2. Sampling Frequency and the Arithmetic Mean
   —Is the calculation of a mean appropriate for small systems?
   —How many samples are enough?
   —How firm is the technical basis for the present monitoring frequency?
3. Compliance With the Coliform Bacteria MCL
   —Is the two part limit practicable?
   —Should follow-up (check) samples be used in calculation of the mean?
   —What is the significance of two negative follow-up samples?
   —Is the MCL properly established on a technical basis?
   —Can the limit be met for *all* systems?

In conclusion, the Interim Primary Regulations for microbiology are basically the same as those contained in the 1962 Public Health Service Standards with certain minor clarifications. These Regulations are adequate, in my opinion, to protect the public health providing the water comes from a protected, uncontaminated source. There are certain desirable modifications that can be made by amendment, others, however, must await further study. Hopefully this symposium will provide guidance to the Agency on how these modifications should be made.

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QUESTION AND ANSWER SESSION

W.B. Jeffcoat, Alabama Department of Public Health, Montgomery, Alabama.

Your statement in session 3, concerning the three problem areas with the regulation is quite true. We in Alabama have relayed our concern to Region IV on the same matters. You only addressed one of these areas, however. The numerical value of "TNTC" and the dual MCL's were not mentioned. You stated that help had been sought to solve these problems. *Help* from where? EPA? States? Universities? Water Systems? Did the help come from people who are *familiar* with the operation of a water supply system or did it come from EPA officials and University personnel whose only contact with a water system is an occasional visit.

I'm sure you realize that federal agencies especially EPA have low levels of credibility. This is something we wish to avoid on the State level. Forcing a water supply to comply with unrealistic requirements will place the State in the same category of EPA. The goal of the SDWA is "safe drinking water for all" and to accomplish this goal cooperation is needed between the State and water system. There is more to operating a water system than merely complying with the standards. I agree with Mr. Taylor and Mr. Harrison that the mean should not be used in determining public notifications. Check samples should also be counted as they do with other parameters in the National Interim Primary Drinking Water Regulations. My main question is will EPA consider the proposals of the utilities and States or will EPA rely on information provided by those unfamiliar with the *actual*, daily operation of public water systems and with the actual, daily operation of a state public water supply supervision program?

C.W. Hendricks, Office of Drinking Water, U.S. Environmental Protection Agency, Washington, D.C.

This meeting was designed to provide the U.S. Environmental Protection Agency with the best information that experts in Water Supply from EPA, the States, Universities and water utilities can provide. I do not believe that any one person, or profession for that matter, has all of the information at hand needed to correct the deficiencies that we see in the coliform maximum contaminant levels. I would anticipate that any amendments to the National Interim Primary Regulations would be thoroughly reviewed by State and utility personnel before they are promulgated.

G.W. Fuhs, New York Department of Health, Albany, New York.

How did you determine there was no danger connected with coliform standard?

C.W. Hendricks.

I would refer you to Mr. Floyd Taylor's paper elsewhere in this manual for a description of how our present coliform standard originated. I suspect that the standards were empirically derived without any study to determine what the risk to health might be. I might add that experience has shown us that the present coliform standards adequately protect the public health providing a high quality uncontaminated source water is used. Any substantive changes in the MCL will require careful evaluation before they are promulgated.

Comment by E.E. Geldreich, Water Supply Research Laboratory/MERL, U.S. Environmental Protection Agency, Cincinnati, Ohio.

If I may, I would like to respond to this presentation by Dr. Hendricks in a series of short comments on the many aspects of the subject.

a. *Two Classes of Water Quality*

The proposed amendments in the bacteriological regulations are drifting into a direction of establishing two classes of public water quality in the Nation with a disregard for identifying poor quality waters in smaller systems. While this may ease the burden of small water supply surveillance on the State agencies, the proposed changes will weaken the goal of achieving uniform, high quality water through minimal regulations for all public water supply systems. Furthermore, these relaxed regulations would be applied to the very water systems that our 1969 national community water supply survey indicated were most subject to producing marginal quality potable water.

b. *Variance in MF-MPN Test Application*

As the regulations now stand there is a variance in the application of rules related to the membrane filter versus the multiple tube test and an uneven interpretation of bacteriological results when using either of these two alternative bacteriological procedures.

Since either of these procedures (MF or MPN) may be used in the examination of potable water, all results should be reporting MF results in terms of coliforms per 100 ml. The present practice of reporting MF results in terms of coliforms per 100 ml while restricting MPN reports to only the number of positive tubes, results in some inequities of interpretation. Every laboratory that is capable of performing the multiple tube test is also capable of computing the proper MPN value. The use of a system that reports only the number of positive tubes per test is archaic, and easily subject to manipulation to meet a monthly minimum and should be discarded.

c. *Recognition of Low-Level Contamination Occurrences*

The definition of an unsatisfactory sample should be "1 per 100 ml" for the membrane filter test and "2.2 per 100 ml" for the multiple tube test. These limits are equivalent since the MPN test value is a statistical estimate and uses a table of numbers elevated by incorporation of a positive bias. This revision in the definition of an unsatisfactory sample would direct attention to the occurrence of persistent low level contamination problems that have been ignored in the past. This change would not alter the intent of the proposed amendment as it relates to repeat sampling since the term "negative" is interpreted to mean that no coliforms were detected in the official test portions by either the MPN or MF procedure. The change would require mandatory check samples for all detected occurrences of coliforms in public water supplies.

d. *Sample Frequency for Small Supplies*

The proposed reduction of sampling frequency for small community systems serving 25 to 1000 persons to one sample per quarter and further reducing for non-community systems of one per quarter on the basis of a sanitary survey should not be permitted. Since the number of supplies serving 25 to 100 persons is largely unknown and these supplies are mostly comprised of small trailer parks and roadside facilities that serve a transient population, sampling should only be reduced to once each quarter during non-recreational periods of the year, however, increased to once per month during the normal recreation period.

Since the burden of increased monitoring and corrective action will be on public water supplies serving populations between 25 and 9,400, guideline dates should be established to intensify program direction first to supplies serving 1,000 to 9,400 people, then to supplies in the 100 to 1,000 population category, leaving the smallest supplies (25 to 100 population served) as the last group to be phased into the program.

e. *Check Sample Incentives*

Check samples should be included in the monthly average of routine samples for all water supplies, regardless of size. This would offer incentive to water plant management to increase their sampling program beyond the minimum two consecutive daily check samples required following an unsatisfactory sample report. Because of the urgency to detect contamination breakthroughs into the potable water, requests for a repeat sampling following unsatisfactory results should be made promptly and the sampling initiated within no more than 48 hours. The answer to the perennial concern about the integrity of the sampling during collection could best be resolved by stating a requirement that sample collectors must be certified by the appropriate state agency following successful completion of a 1-2 day training course. Thereafter, recertification could be related to observed performance of sample collectors during the recertification of the laboratory.

f. *Public Notification*

The public notification requirements should be restructured to include two levels of response. The first level of response occurs when one to four coliforms are detected by the MF method or 2.2 to 5.1 coliforms are reported in the MPN procedure. In this case, the water system must institute an immediate review of treatment practices, including examination and cleaning (if necessary) of the distribution system. Daily check samples are mandatory during both the sanitary survey and the period of corrective action until two consecutive satisfactory samples are obtained. A written record of the survey report, corrective action(s) and bacteriological results are maintained for five years. The second level of response involves any occurrences of 5 or more coliforms per 100 ml on the MF or above 5.1 per 100 ml as measured by the MPN test in one or more of the daily check samples. In this later situation, public notification would be mandatory, including a "boil water order," if necessary, immediate increases in disinfection to achieve a disinfection residual in the most remote sections of the system, hourly measurements of disinfectant residuals and daily bacteriological sampling at these remote sections. As soon as the monitoring data on the distribution network demonstrates a disinfectant residual for 24 continuous hours, and two consecutive days of satisfactory bacteriological results, a public notice cancelling the "boil water order" should be issued.

g. *Program Approach*

In essence, while portions of the regulations need to be changed to resolve problems in monitoring and interpretation of laboratory results, these modifications must not drift toward establishing two classes of public water quality in the Nation with a disregard for identifying and responding to poor quality waters in smaller systems. A systematic approach to attacking the increased program responsibility should be geared to solving current constraints in state and local water supply staffing and monitoring costs, not in minimizing objectives defined by intent of the law.

Experiences With the Coliform Standard Under the Interstate Carrier Program

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Introduction

The past often provides the record of experience which is useful in guiding decisions on present and future courses of action. History is also littered with the wreckage of men and systems who ignored its lessons and this of course is what we seek to avoid by the intensive examinations we are giving this "Evaluation of the Microbiology Standards for Drinking Water." Our particular part of this examination deals with experience with the coliform standard under the interstate carrier certification program conducted for 56 years by the U.S. Public Health Service and for seven by the Environmental Protection Agency. I believe it will be helpful to briefly review the development of the coliform standard as it appeared in successive issues of the U.S.P.H.S. Drinking Water Standards.

The Coliform Standard

In 1914 the entire document, commonly referred to as the Treasury Standards, was entitled "Bacteriological Standard for Drinking Water," under which Secretary McAdoo informed the Surgeon General that "In the future common carriers will be required to furnish water for passengers in interstate traffic which will conform to this standard." (5) The sections of that standard pertinent to today's discussion read as follows: "Not more than one out of five 10cc portions of any sample examined shall show the presence of organisms of the *Bacillus coli* group when tested as follows:" and "The total number of bacteria developing on standard agar plates, incubated 24 hours at 37°C shall not exceed 100 per cubic centimeter." (6) It should be noted that no mention was made of the number of samples to be taken nor of the deviations from the above stated requirements which would appear in later revisions. The next Treasury Department document was entitled "Drinking Water Standards" and we see the beginning of the present day practice for it read, in part, as follows:

"II. AS TO BACTERIOLOGICAL QUALITY

"1. Of all the standard (10cc) portions examined in accordance with the procedure specified below, not more than 10 percent shall show the presence of organisms of the *E. coli* group.

"2. Occasionally three or more of the five equal (10cc) portions constituting a single standard sample may show the presence of *E. coli*. This shall not be allowable if it occurs in more than

(a) Five percent of the standard samples when 20 or more samples have been examined;

(b) One standard sample when less than 20 samples have been examined.

Note—It is to be understood that in the examination of any water supply the series of samples must conform to both the above requirements 1 and 2. For example, where the total number of samples is less than six, the occurrence of positive tests in three or more of the five portions of any single sample, although it would be permitted under requirement 2, would constitute a failure to meet requirement 1."

In the 1942 and 1946 document by then called the Public Health Service Drinking Water Standards—standard portions and standard samples were defined and the 100 ml standard sample was introduced as were the first requirements as to the number of samples which should be collected. The well known graph showing the relation between population density and number of samples to be collected per month appeared in the 1942 standards. It will be helpful to note the slight changes in the allowed limits as they have bearing on today's discussion. The requirement was as follows:

"12.203(b)(1) of all the standard ten milliliter (10 ml) portions examined per month in accordance with the specified procedure, not more than ten (10) percent shall show the presence of organisms of the coliform group."

"12.203(b)(2) occasionally three (3) or more of the five (5) equal ten milliliter (10 ml) portions constituting a single standard sample may show the presence of organisms of the coliform group, provided that this shall not be allowable if it occurs in consecutive samples or in more than

(i) Five (5) percent of the standard samples when twenty (20) or more samples have been examined per month.

(ii) One (1) standard sample when less than twenty (20) samples have been examined per month.

Provided further that when three or more of the five equal ten milliliter (10 ml) portions constituting a single standard sample show the presence of organisms of the coliform group, daily samples from the same sampling point shall be collected promptly and examined until the results obtained from at least two consecutive samples show the water to be of satisfactory quality." It is not necessary to emphasize the requirements for 100 ml sampling as this system has not been widely adopted even though it has been carried over into the EPA Interim Primary Drinking Water Regulations. It will be realized that the 10 ml multiple tube series for the 1925 and 1942 standards were specifying an average of less than 2.2 coliform organisms per 100 ml with occasional allowances as high as over 16 organisms per 100 ml, but, beginning in 1942, only if check samples (two consecutive) were negative. In other words a high sample was the signal to take corrective action.

In the last (1962) PHS Standards the general wording of the 1946 edition was continued but with the further restriction that "the presence of the coliform group in three or more 10 ml portions shall not be allowable if this occurs in two consecutive samples" (2). Also in 1962 the membrane filter technique was recognized with the requirements not to exceed an average of one colony per 100 ml (monthly arithmetic) and on occasion not more than 4/100 ml. And that is how we arrived where we are today with the bacteriological requirements of the Interim Primary Drinking Water Standards which are essentially those of the 1962 PHS Standards. I would however point out that the 1914 document and its successors, or rather the application of them together with the practice of filtration and disinfection were responsible for the dramatic reduction of enteric disease in this country during the early 1900's.

Coverage of the Standards

When the first federal drinking water standards were promulgated there were in 1916, 4077 U.S. public water supplies which came under their jurisdiction. It is estimated that they then served about 35% of Americans who utilized public water supplies. In 1975 the number of Interstate Carrier Water Supplies was 689 serving 90,000,000 people or about 55% of the population on public water supplies. I would also cite the historic policy by which the standards were applied namely through a cooperative, federal-state program. Even more importantly, the standards were adapted by nearly all the states as applicable to public water supplies generally within their jurisdictions. However, judging from experience in New England, this application was administratively and not legally enforced. Now a word about the general procedures for applying the bacteriological standards.

Application of Standards

Records of the precise methods of applying the first drinking water standards were difficult to find but Treasury Department Circular No. 48, October 5, 1912 directed PHS commissioned officers traveling on common carriers to make observations regarding sanitary conditions and submit appropriate reports (1). More importantly, the states were required to make reports to the Service on the quality of the sources and presumably gave warning when the quality of a supply failed to meet the requirements. Let me here state for the benefit of those who are not aware of how the Interstate Carrier Water System worked, the enforcement phase has always been an action between the federal government and the common carrier not between the federal government and the water purveyor. Furthermore, since the early days of the system the states made recommendations as to the type of action which should be taken.

Sometime between 1925 and 1938, when my personal experience with the system began, a three level method for approving carrier water supplies was adopted. First there was *full approval*, second *provisional approval* and third, *prohibited*. The first meant that all was well, the second that there was some question about the capability of the supply to continuously provide safe water and prohibited meant what it said. What the classification would be was somewhat arbitrarily determined by discussions between the state health engineer and the PHS district engineer.

Beginning in 1942 the concept of numbers of samples per month and allowable ratios of coliforms were adopted and an annual report form was introduced which required the State to show for each month what the bacteriological record had been. However these reports were submitted only once a year to PHS and "bad" results (which by that time could be a year or more old) were usually explained by a footnote. Invariably the supply was, at worst, "provisionally approved." With the great shift during this period of personnel and resources to water pollution control activities at both state and federal levels, interest in water supply declined almost proportionately. By 1959 many state and Federal water supply programs had fallen into a state of low effectiveness. In that year the Public Health Service water supply program was placed under new management.

In 1962 Public Health Service Headquarters Water Supply Program advised all the regional offices that the bacteriological requirements * of the standards meant what they said and would be enforced. In the ensuing years the numbers of provisional and prohibited actions increased markedly. It was also during those years that the Service began to keep track of the water supply improvements that resulted from all enforcement actions and although I cannot quote you exact data I can tell you that the rate of new water supply construction resulting from the program was of the order of several millions of dollars per year.

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* HEW General Counsel determined that only the bacteriological provisions of the standards were enforceable.

In 1971 when the Interstate Carrier Water Supply Program had gone over to EPA the first systematic guide to the Interstate Carrier Water Supply Certification Program appeared entitled "A Guide to the Interstate Carrier Water Supply Certification Program (7). It provided valuable criteria for certifying Interstate Carrier Water Supplies including both bacteriological and chemical. Again the former are of particular interest to us in this discussion and the basic elements were as follows:

For Approved:

Water quality must meet all the limits of Section 3.2 (sections as they appear in the 1962 standards) and must meet the sampling ratios of Section 3.1 at least 11 out of each 12 month period.

For Provisional Approval:

Water quality failed to meet Section 3.2 requirements for one of any 12 month reporting period and/or monitoring requirements were failed for 2 or more months.

Use Prohibited:

Water quality failed to meet Section 3.2 requirements for 2 or more months of any 12 month period and/or monitoring requirements were violated by failure to obtain less than 50% of the required number of samples for 3 months of any 12 month period.

To get back to a "provisional" from a "prohibited" classification the supply must have a record of at least three consecutive months of satisfactory bacteriological results and sampling frequency.

I can not speak for the impact of the systematized requirements outside of Region I but I can say that there it has resulted in better reporting, and upgrading of monitoring frequency and the improvement of plants and operations. In one state we have seen an improvement from 1970 when 70% of the public water supplies did not meet bacteriological standards to 1976 when 50% did meet them.

I would also like to cite two cases in point where failure to meet the bacteriological requirements of the Drinking Water Standards resulted in prohibited actions which encouraged needed improvements. The first was Springfield, Massachusetts and the second Fall River, Massachusetts. Until about 1969 Springfield, a city of some 160,000 provided water with slow sand filtration as the only treatment. Frequent instances of coliform levels in excess of the PHS standards had been noted and a sanitary survey revealed less than adequate watershed protection. The city was requested to, and did, install a $125,000 automatic, chlorination station since which installation the bacteriological record has been excellent.

In the summer of 1969 Fall River experienced widespread contamination of its distribution system and the state issued boiled water orders to its 100,000 inhabitants. The PHS issued a "Prohibited Use" certification and refused to change the classification until adequate treatment was assured. Today Fall River is using a modern water filter plant which cost some $4.5 million most of which was paid by a HUD grant.

Conclusions

I believe I have complied with my instructions to tell you how the bacteriological limits of the interstate carrier standards were implemented from Public Health Service Headquarters and regional offices and what were some of the results. The third question is more difficult. "Should we continue to implement the limit for communities in the same way as indicated in the interim regulations: my recommendation is "yes" as far as the numbers of allowable coliforms are concerned and "no" as concerning the monitoring and public notification requirements.

Let me next say that some of you would have recognized a similarity between the "provisional approval" of the old ICWS system and the variance and exemption provisions of the SDWA. These allowed room or time in which to negotiate. I firmly believe this room to negotiate plus the Use Prohibited action clause, where needed, accounted for our success under the ICWS program. These principles should continue to govern our program.

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Experiences with the Coliform Standard

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Introduction

When Dr. Charles Hendricks invited me—almost two months ago—to participate in this symposium regarding standards for bacteriological quality of drinking water, I in turn asked each of the nine other EPA Regions and a number of States to help me gather the facts and figures that would document experiences with the historic coliform standard. I have received responses from most Regions and about eleven different State agencies.

I will describe some typical experiences that public water systems have in finding coliform bacteria, and how various State agencies and EPA regions have treated these findings in the past. There has been more diversity than consistency in the supervision of bacteriological quality. But I think you will agree that no State or EPA Region has ever been as stringent as the National Interim Primary Drinking Water Regulations (NIPDWR) are now asking us to be.

It is not fair for me to take the entire credit for this paper as it is almost entirely pieced together from the inputs of others. A number of people have written me case history descriptions of bacteriological problems including follow-up findings and actions. I will quote a number of them directly. The cases illustrate some important points and I am sure you will enjoy them as I did.

I guess one reason that I am here today is that I have some very strong feeling about the current microbiological standard. It is not logical; water systems cannot comply with it; and it cannot be enforced as currently written in 40 CFR 141.14 and 141.21.

Coliform bacteria in drinking water represent an acute rather than a long term health effect. A single swallow of coliform is just as dangerous as a monthly average exposure. The bacteriological safety of drinking water should not be defined by an arithmetic mean of all samples or percent of positive portions over a month's period. The individual sample limits define the *safety* of drinking *water*, whereas a monthly or yearly average coliform limit pertains to the *reliability* of drinking water *systems*. By applying both limits, as we have in the December 24 NIPDWR, we end up with a double standard. Public water system consumers can, according to our current standards, safely drink water with four coliforms per 100 milliliters every day of the month until the last day when this same water suddenly becomes unsafe because the computed monthly average exceeds one coliform per 100 ml.

It is a good idea to use long term averages or percentages of bacteriological data to judge the adequacy of water system facilities and operations. But I am absolutely convinced we must use only the individual sample limits to define safe drinking water. Monthly summaries of bacteriological data should do no more than trigger investigative action to ascetrain that a serious health hazard is not lurking on the sideline and waiting to emerge. It is not proper to define water with four coliforms per 100 milliliters as safe to drink and at the same time say water with more than one coliform is unsafe.

The second major flaw regards public notification. The December 24 regulations require that the public be notified on the basis of unverified bac-

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teriological data. In fact, as Section 141.21(f) is currently written, a single unsatisfactory bacteriological sample result will force nearly all public water systems in this country to give public notice.

P.L. 93-523 public notification respecting acute health effect contaminants such as bacteria and nitrates is *not* meant as a warning for consumers to take emergency action in their own behalf. States will still have to issue discretionary boil orders and other emergency advisories for this purpose, independent of SDWA requirements.

The Safe Drinking Water Act (SDWA) notice, on the other hand, will be included in the water bill and is rather a more casual news letter and historical record of a past water quality problem, the problem's significance, and what is being done about it. The State implementation regulations say that a State qualifying for primary enforcement responsibility need only require notice in the water bills or, if the bills are less frequent than quarterly, by another form of direct mail. The water bill announcements will be at least a week or perhaps months later than the occurrence of a bacteriological problem. This notice should further include "a balanced explanation of the significance or seriousness to the public health, a fair explanation of steps taken to correct any problem, and the results of any additional sampling." This is all in keeping with the intent expressed in House Report 93-1185, which states: "The purpose of this notice requirement is to educate the public as to the extent to which public water systems serving them are performing . . ., to develop public awareness of the problems facing public water systems, and to encourage a willingness to support greater expenditure at all levels of government to assist in solving these problems."

It is important therefore, that these information notices be factual and valid. We must be certain they are not triggered by spurious results that would unduly alarm the public. There is, in fact, good argument for public notice to be determined only after the results of a compreheisive investigation. But with the inherent error we will always have in microbiological testing, the SDWA violations and public notice should *NEVER* be based on the result of a single coliform test. If we ever start enforcing on the basis of non-valid results and no actual health hazard, water system operators will soon begin to bias samples toward negative results rather than search for water quality problems.

I will explain my thoughts and recommendations for correcting these anomalies in a while. But first I would like to relate some specific experiences with the coliform standard that a number of people have helped me document for this symposium.

EPA Regions' Experience With the Coliform Standard

A guide to the Interstate Carrier Water Supply Certification Program was written in 1971 by EPA's water supply program. It was, perhaps, the first attempt to establish uniform criteria and procedures for interpreting the drinking water standards (DWS) and for classifying interstate carrier water supplies (ICWS). But even with the use of this uniform guide, the 20% of ICWS recorded in the latest tally as "Provisionally Approved" does not reflect the total number of bacteriological discrepancies that were found nor the number that would have failed the present microbiological MCL (maximum contaminant level). Professional judgment can never be completely eliminated from evaluation of the bacteriological safety and reliability of public water systems.

Region III gave two good examples of where judgment was wisely used to override a classification that the numbers alone would have dictated.

Chesapeake City, Virginia—50,000 population served

This supply collects between 70 and 100 bacteriological samples per month from its distribution system and performs the MF analysis. In July of 1976 they collected 92 samples. That month, one bad sample was found with a coliform density of 520 per 100 ml. This one sample made the mean density for that month calculate out to 5.7 per 100 ml and thus exceed the standard. Appropriate check samples were collected and they came out negative. It is a very good possibility that there was a sampling error. This should have resulted in an ICWS downgrade to a "Provisional" classification. In our judgment there was no health hazard present and no action was taken to downgrade the system.

The December 24 NIPDWR are written—even with the latest changes we heard about today—to take this latitude away on June 24.

Another case involves the Philadelphia Surban Water Company—900,000 population served. Region III accepted the explanation given in the following letter without any downgrade in classification.

Mr. Walter Stanley
Pennsylvania Dept. of Environmental Resources
1875 New Hope Street
Norristown, PA 19401

Dear Mr. Stanley:

This will explain, for the record, why we reported a mean coliform density of 2.3 organisms/100 ml for the week ending July 28, 1973.

During that week a number of samples from our system showed the presence of coliform organisms. However, we began to doubt the veracity of the results since they reflected points widely scattered throughout our several divisions (300 square mile area).

A thorough investigation made early in August showed that the "bad" samples were not due to the source—but to one particular sample man. We established this fact by having a second man follow the first one and sample the same points for several days. Without exception, the first man's samples showed coliforms, whereas those collected by the follow-up man were negative.

Even a thorough medical examination failed to show how the first sampler's samples became contaminated. Nevertheless, we took this man off the sampling run and gave him thorough training in bacteriological sampling technique. He is now back on the job and everything appears to be in order.

I don't know what happened, but I am convinced that the reported coliform count was due to sampling technique and not to any impaired quality of the water.

Sincerely,
Kenneth E. Shull

The Philadelphia Suburban Water Company affords better quality control and testing than most public water systems covered by the SDWA will ever obtain. The example points out that even the best laboratory certification and quality assurance program cannot hope to eliminate all imprecision and error from microbiological testing. Margins of error must be allowed in determining compliance with the MCL, and professional judgment must be allowed in determining the degree of health risk and appropriate follow-up when bacteriological monitoring raises a question about the reliability of water system facilities or operation.

Region IV uses a case history concerning Florence, South Carolina, to point out two key weaknesses in the microbiological standard:

> On February 24, 1972, the Region was advised by the State that the Florence System had had eleven highly contaminated samples on February 14 and 15, such that they would surely fail for the month. At that time, it also became known that the system had failed in October 1971, based on one sample out of 36 unsatisfactory with a count of 47 for a mean coliform density of 1.3. Subsequent sampling (not rigorous check sampling) showed no problems. After a joint USEPA-State investigation, the system was classified USE PROHIBITED on February, with appropriate telegrams and a press release.
>
> This instance illustrates two problems with the Microbiological MCL: (1) One single unsatisfactory sample in October caused the system to fail and would have necessitated public notice under the Interim Primary Drinking Water Regulations; (2) Official action on the flagrantly bad quality detected in mid-February was not warranted until the end of the month, when problems had disappeared.

It should be noted in this instance that although the Florence system was classified "Use Prohibited", the State gave public assurances of no health danger. State agencies in the past have not rigorously determined compliance with bacteriological standards. Even though State standards are very similar or identical to the PHS standards, enforcement actions are only contemplated when contamanition is substantially more serious than that minimally necessary to fail the MCL. Many States feel the real value of the bacteriological test is to paint a monthly and yearly picture of water quality which can be considered, along with knowledge of the system and its operation, to reach a conclusion as to whether that system is adequate. Some States do require resamples whenever routine results indicate that coliform is present. But the most effective follow-up is that provided through direct one-on-one contact between the field engineer and the water supply. This follow-up is by and large at the discretion of the engineer. One such person in Region V who, I know, is doing an outstanding job gave me this description of his operation:

> It is my policy that when one sample has a count of 10/100 ml or more, or two or more samples show contamination, to call the results to the supply operator. At this time we attempt to determine if anyhing has happened which might have caused the contamination. Based on our telephone conversations, we make specific recommendations such as: boil orders, increased chlorine residuals, collect samples from proper taps, resampling, etc. Recommendations obviously vary considerably depending upon information obtained.

I would not want to see this work interrupted for the purpose of enforcing additional monitoring, reporting, and public notification where water quality is not impaired or a health hazard does not exist.

One of the most striking statements I received was written by Leroy Stratton of the Illinois Department of Public Health. He noted that facilities which would be regulated by his Department "are primarily non-community supplies with none exceeding 1,000 population." These systems will be required to sample on a quarterly basis, therefore "requiring public notice any time a contaminated sample is received, regardless of the level of contamination." I would add that two-thirds or more of the nation's community water systems are also less than 1,000 population, likely to be sampling once every three months, and therefore require public notice every time a contaminated sample is received.

Don Taylor, in EPA's Region IV office, examined 12 months of records for 122 public water supplies and reached these conclusions based on the December 24 regulations:

"1. The "average" public water supply will fail the monitoring requirements almost 5 (4.77) months each year.
2. The "average" public water supply will fail the bacteriological quality provisions approximately one month in each 21 months of record.
3. The "average" public water supply would go to public notice approximately 5 or 6 times per year."

One final example again from Region IV points out the fallacy of the monthly average limit for dictating water quality safety.

An interstate carrier water system, Ingalls Shipbuilding-West in Pascagoula, Mississippi, showed chronic inability to consistently maintain bacteriological quality in 1972 and 1973. Strict application of the PHS Drinking Water Standards and the "Guide to the Interstate Water Supply Certification Program" resulted in the following sequence of actions:

3/10/72—System classified USE PROHIBITED (with press release) based on failure to meet standards 11/71 and 2/72.
5/11/72—System reclassified PROVISIONALLY APPROVED (with press release) based on 3 months good record.
2/15/73—System classified USE PROHIBITED (with press release) based on failure in 1/73.
8/ 1/73—System reclassified PROVISIONALLY APPROVED based on 4 months good record. (System also failed in 2/73).
9/14/73—System classified USE PROHIBITED (with press release) based on failure in 8/73.
1/17/74—System reclassified PROVISIONALLY APPROVED based on 4 months good record.
9/19/74—System reclassified APPROVED based on 12 months good record.

The classification actions outlined above resulted from five individual months of failure to meet standards. In only one of these months was the failure the result of a single unsatisfactory sample (1 unsatisfactory out of 4). In the other four instances, at least two samples were unsatisfactory.

This example points out the problems of small systems meeting the standards, even with onus of unfavorable publicity.

The official record includes another noteworthy month. In August 1972, 83 samples were collected, one was unsatisfactory with a count of 80. During the previous and subsequent months four samples had been collected monthly. The unsatisfactory sample would have meant another USE PROHIBITED situation, and evidently approximately 79 additional samples were taken to bring the system into compliance.

The best way to eliminate the monthly average dilemma is to eliminate the monthly average limit. A better monthly summary is simply the number or percentage of positive coliform results in any one month. But an important distinction must be made between any monthly summary coliform limit and an individual sample limit. The first pertains to the *reliability* of water *systems*, and the second pertains to the *safety* of drinking *water*.

The occurrence or recurrence of any coliform in drinking water should be investigated to ascertain and correct potential health hazards. But only one limit should actually define safe drinking water. We must remember that unreliable water systems can produce safe drinking water, and discrepancies respecting a low monthly summary limit may be only a potential rather than an actual or existing health hazard.

Proposals

As stated in the preamble to the December 24 National Interim Primary Drinking Water Regulations, "further changes in the microbiological regulation may be desirable." A number of people other than myself have given this considerable thought, and have had input into the following two proposals. These two alternatives I feel are logical. Either one, if chosen, is practical enough to allow implementation.

The text of Section 141.14 and parts (d), (e), (f), and (g) of Section 141.21 in the December 24 regulations should be deleted. Part (h) of Section 141.21 should be labeled (g). The following then inserted as replacement and change:

> Section 141.14
>
> The following are the maximum contaminant levels for coliform bacteria applicable to community water systems and non-community water systems. Compliance with maximum contaminant levels for coliform bacteria is determined pursuant to Section 141.21.

The last sentence is consistent with the MCL regulations for organic and inorganic chemicals. It allows compliance to be defined by follow-up activities, and it prevents causing violations and public notice on the basis of a single sample result.

PROPOSAL 1

> Section 141.14 (Cont'd)
>
> (A) When the membrane filter technique pursuant to Section 141.21(a) is used, the number of coliform bacteria shall be less than one per 100 milliliters in any sample collected and analyzed pursuant to 141.21(b) or (c).
>
> (B) When the fermentation tube method and either ten milliliter standard portions or 100 milliliter standard portions pursuant to Section 141.21(a) are used, coliform bacteria shall not be present in any portions in any sample collected and analyzed pursuant to Section 141.21(b) or (c).

Bacteria in drinking water represent an acute health effect and the presence of any coliform organism in finished drinking water suggests either inadequate treatment or access of undesirable materials to the water after treatment. Coliform also may be indicative of dangerous contamination and of water that is not safe to drink. The limit therefore simply pertains to the presence of any coliform in drinking water. It eliminates the need for an arbitrary allowable number and it is easily understood.

> Section 141.21
>
> (d)(1) When a sample exceeds a maximum contaminant level set forth in Section 141.41(a) or (b), one check sample shall be collected from the same sampling point and examined. If a subsequent sample has already been taken from the sampling point, it shall be considered the check sample.

If a supply monitors regularly as prescribed in (b) and (c) of this section and there does not appear to be serious contamination, then one check sample should be enough. However, paragraph (3) will allow the State to require additional check samples if a health hazard is suspected. Counting the next subsequent sample as the check sample makes a great deal of sense but wasn't spelled out in the December 24 regulations. Many water systems will have already resampled from the same tap before the results of the previous sample are known.

(2) When the examination of the check sample required in Section 141.21(d)(1) shows the presence of coliform organisms, the supplier of water shall:

(I) Report to the State within 48 hours;

(II) Initiate an investigation, including the collection and examination of additional check samples from the same point and other sampling points in the area, to define the extent of the problem; and

(III) Notify the public in the area affected by the indicated contamination as prescribed in Section 141.32 or the State public notification requirements, unless the State determines that no health hazard has actually existed.

Based on the results of the check sample, the supplier of water must 1) report to the State, 2) initiate an investigation, and 3) notify the public. An important provision regards public notice requirements only *to the area affected by the indicated contamination*. For large regional water systems it is folly to require notification to perhaps one-half the State's population when the problem may be limited to one section of one town, one water main, one building, or one faucet. Of course, the utilities would have the option to notify all consumers if they so choose.

(3) The State, at its discretion, may require that additional check samples be collected at a specified frequency from the same sampling point and other sampling points in the area and examined to identify and eliminate suspected health hazards when a sample exceeds a maximum contaminant level pursuant to Section 141.14(a) or (b), even if the check sample required in Section 141.21(d)(1) does not indicate the presence of coliform bacteria.

Some sample results or bacteriological histories may raise a question about the reliability of the system to continuously supply safe drinking water even though no unsafe water has yet been found. In this case the State may require additional samples to identify and eliminate suspected health hazards.

(4) When the cause of the indicated contamination has been determined and corrected, additional check samples shall be collected at a frequency directed by the State.

Just to confirm that the correction has been effected, additional samples may be wanted for a specified time period.

(5) The location at which the check sample was taken pursuant to Paragraph (d)(1) of this Section shall not be eliminated from future sampling without approval of the State.

Same requirement as in the December 24 regulation.

(E) The State may determine that unreliable examination results for a sample collected in a discreet monitoring period pursuant to Sections 141.21(b) or (c) were caused by factors beyond the control of the water supplier. Such factors could be excessive transit time between collection and examination of the sample, samples being broken in transit, or interference test results when the membrane filter technique is used. If this is the case, another sample collected immediately upon learning

TABLE 1. EXPERIENCE WITH THE COLIFORM STANDARD.

	Number of Annual Public Water System (PWS) Records Reviewed	1-4 coliforms per 100 ml.		> 4 coliform per 100 ml.	
		Percent of PWS with 1-4 coliform per 100 ml at least once/yr.	Percent of occurrences found on follow-up to be a health hazard	Percent of PWS with >4 coliform per 100 ml at least once/yr.	Percent of occurrences found on follow-up to be a health hazard
Ilinois					
IEPA	326	8.6%	zero	11.6%	33.9%
IDPH	988	2.6%	zero	28.1%	4.3%
Indiana	550	12.1%	—	—	—
Wisconsin	93	30.1%	—	6.4%	—
Region V					
ICWS	306	6.8%-11.8%	—	6.7%	—
Region II					
ICWS	400	—	—	3.8%	—
Oregon	1,118	5.4%	—	10.6%	—
Region IV					
ICWS	550	11.5%	—	7.2%	—
New Mexico	152	7.2%	—	15.8%	—
Texas	3,347	0.8%-3.1%	—	3.2%	—
Total	7,830				
Average		10%	zero	10%	20%

> of these results may be used in determining compliance with sampling requirements in Section 141.21(b) or (c). However, a single sample may not be attributed to more than one monitoring period.

Overgrowths, "too long in transits"—over 48 hours not 30 hours, and broken bottles frequently account for 10% of the samples being invalid or the results unreliable. These samples should be disregarded, but unless the operator is then allowed to collect and count another sample, he will be in violation of the monitoring requirement through no fault of his own.

> (F) Check samples, samples with unreliable examination results, and special purpose samples, such as those taken to determine whether disinfection practices following water main placement, replacement, or repair have been sufficient, shall not be used to determine compliance with Section 141.21(b) or (c).

Same as in the December 24 regulation, except samples with unreliable examination results are not counted as well.

Table 1 was compiled with data from 10 different Regional and State agencies. It shows that ten percent of public water systems experience one to four coliform organisms per 100 milliliters at least once per year. Seldom have these occurrences been found to present a health risk. Another ten percent of public water systems experience over four coliforms per 100 milliliters at least once per year and twenty percent of those occurrences have been found on follow-up to be of significance.

These data point out that a limit of one coliform may be more regulation and more expense than is needed to *define* safe drinking water. Federal drinking water standards have for over 50 years defined water with four coliforms per 100 milliliters as safe to drink and there is no evidence now saying it is not. Some State agencies from a practical standpoint may want to or have to prioritize follow-up regarding bacteriological results. This latitude to four has historically existed in the Public Health Service standards and may be needed in the future when SDWA requirements are applied to hundreds of thousands of small public water systems. Proposal 2 divides the standard into two parts—141.14(a) and (b) addresses the *safety* of drinking *water* and 141.14(c) addresses the *reliability* of drinking water *systems*. The first paragraph of Section 141.14 and changes to Section 141.21 would remain as I have discussed with Propsal I.

PROPOSAL 2

(Section 141.14 cont'd)

(A) When the membrane filter technique is used, the number of coliform bacteria shall not exceed four per 100 milliliters in any sample collected and analyzed pursuant to Section 141.21(b) or (c).

(B)(1) When the fermentation tube method and ten milliliter standard portions are used, coliform bacteria shall not be present in three or more portions in any sample collected and analyzed pursuant to Section 141.21(b) or (c).

(B)(2) When the fermentation tube method and 100 milliliter standard portions are used, coliform bacteria shall not be present in five portions in any sample collected and analyzed pursuant to Section 141.21(b) or (c).

(C) The State and public water system shall initiate definitive action to identify the cause of the positive bacteriological sample results and to eliminate potential health hazards which migh exist in the system when monitoring pursuant to Section 141.21(b) or (c) shows the presence of any coliform organisms in either of the following:

(C)(1) more than ten percent of the samples when 20 or more samples are examined per month; or

(C)(2) more than two samples when less than 20 samples are examined per month.

QUESTION AND ANSWER SESSION

C.W. Hendricks, Office of Drinking Water, U.S. Environmental Protection Agency, Washington, D.C.

Do you have any recommendations for a simpler standard of sample collecting to eliminate the confusion caused by terminology of "less" or "more than 20 samples per month"?

J. Harrison, Water Supply Branch, U.S. Environmental Protection Agency, Chicago, Illinois.

My proposal 1 and the first parts of proposal 2 simply refer to the result of any sample, and eliminate confusion caused by different standards for those systems collecting less or more than 20 samples per month. This adequately covers a definition of *safe* drinking *water*. However, if addressing the *reliability* of water *systems*, one must be concerned about repetitive sample results. For those systems collecting 20 or more samples per month, it is best to regulate repetitive results by a percentage of all samples collected; for those collecting less than 20 samples, by a simple number.

M.J. Allen, U.S. Environmental Protection Agency, Cincinnati, Ohio.

Are you defining a "spurious" water sample as one sample which results in raising the monthly average greater than maximum MCL? If not, what is your definition of a "spurious" water sample?

J. Harrison

I used the word "spurious" in the context that Safe Drinking Water Act notices to the public should not be "triggered by spurious results that would unduly alarm the public." By a spurious result I mean a false or illegitimate analytical result that does not reflect quality of the water but is rather caused by an error in analysis, sample collection, transit, or other anomalies not directly related to the water supply system. Such anomalies and false results do occur and they are the reason that nitrate as well as all other maximum contaminant levels, except coliform bacteria, are never violated on the basis of a single unverified sample result. It is not proper that the December 24 regulations allow spurious results to raise the monthly average greater than the MCL, and require over 90 percent of all public water systems—those required to collect only one sample each reporting period—to notify the public every time a positive bacteriological sample result is obtained. My proposals 1 and 2 would both define a microbiological violation and attendant notification on the basis of two—the check sample also being unsatisfactory—analytical results.

Implementation of the Coliform Standard by a Community Water Utility

Robert J. Becker
Indianapolis Water Company
Indianapolis, Indiana 46202

As the sole representative of the public water supply industry, I feel it is appropriate to begin with a disclaimer statement. Namely, that the views and opinions expressed by the speaker do not necessarily represent those of all community water supplies governed by the Safe Drinking Water Act.

I must assume my presence here today reflects the desire of the Environmental Protection Agency to obtain and evaluate the bacteriological operating data from a representative and responsible water utility. Thanks to a number of illustrious predecessors, the Indianapolis Water Company has a continuous record of bacteriological data on raw, treated, and distribution system samples dating back over fifty years.

The Indianapolis Water Company is an investor owned utility serving approximately 700,000 water consumers in Central Indiana. Currently 95% of our annual supply comes from two surface streams which during low flow periods become rather scrawny. Each supply is supplemented from upstream storage impoundments during these low flow periods. These streams receive a significant proportion of municipal wastewater discharge as well as agricultural runoff. While they are undoubtedly not ideal supplies, I feel certain they are far from the worst.

Our treatment facilities would be considered fairly conventional. Coagulation is with alum, and the mixing and settling basins provide a designed retention time of between two and one-half to four hours. Disinfection is achieved with chlorine application prior to mixing and settling at a rate sufficient to maintain a free chlorine residual through the filters. All filters are of the rapid sand type with a six inch anthracite coal cap.

As is the common practice of almost all water utilities in this country we rely almost totally on chlorine to provide a bacteriologically safe water to the consumer's tap. During the winter months chlorine residuals leaving our treatment facilities are maintained slightly in excess of 1.00 ppm. This residual is increased to about 1.50 ppm during the hot summer months. Normally, we apply anhydrous ammonia to the filter effluent to convert to a chloramine residual prior to delivery to the distribution system. We realize that a chloramine residual is a less potent bacteriocide than a free chlorine residual. Therefore, for about a four week period each spring and fall we discontinue ammonia application, and treat the entire system with a free chlorine residual. It is important that this occur while water temperatures are not much greater than 50°F.

As a system supplying a population of approximately 700,000, we are required to collect 250 samples monthly. Actually, we will run about 375 samples per month. Our distribution system sampling points are selected for their strategic location, general sanitary conditions, and availability for our regular entrance. At the present time, we have 53 sample sites located throughout our service area. Due to distances involved to the extremities of the system, we have four samples trips. Each trip involves 15 to 20 samples. Obviously, some locations are sampled more frequently than others. It is our intention to have the preparation of all samples completed within four hours of collection. All of our outlying booster stations and elevated

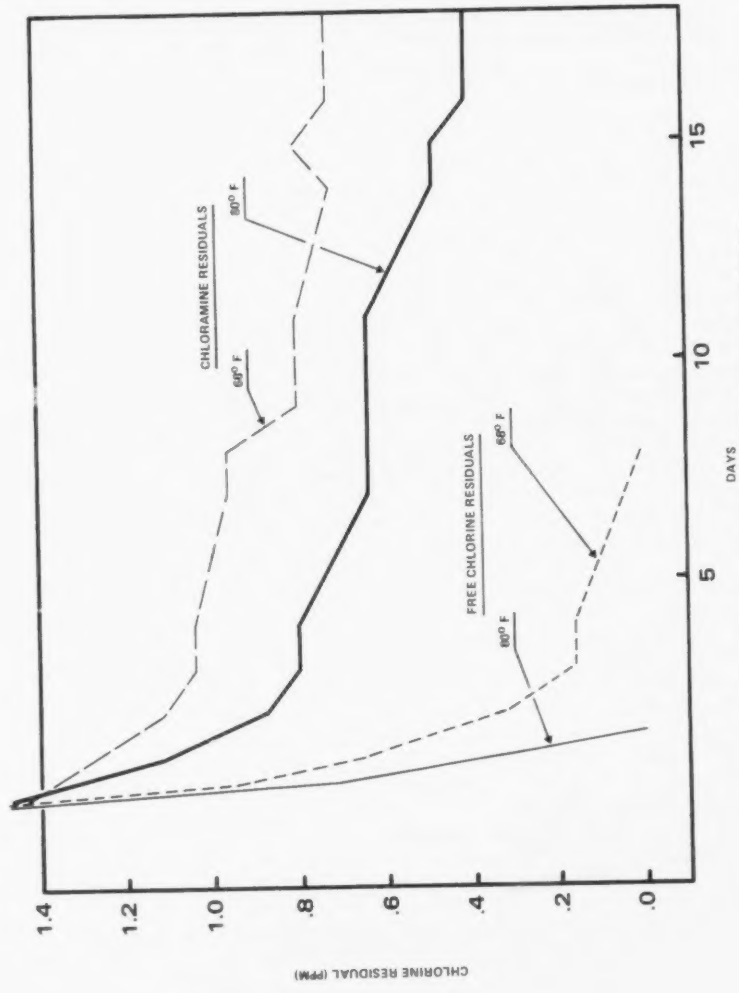

Figure 1. PERSISTENCE OF CHLORINE RESIDUALS AFTER 5 HOUR CONTACT TIME. WHITE RIVER PLANT EFFLUENT.

tanks serve as sampling sites. Other sites include fire stations, filling stations, and the like. We purposely have a few sampling sites on or near dead ends. We are trying to maintain a knowledge of water quality throughout the system, and not just obtain a good record to impress the public or the public health authorities. We do not now have nor ever had during our period of record any open or uncovered finished water reservoirs.

There are many factors which influence the water quality which is delivered to the consumer's tap. One of the great limitations in achieving uniform quality throughout an entire distribution system lies in our inability to maintain adequate levels of chlorine residuals indefinitely. We have documented this occurrence in a series of laboratory experiments. Sterile gallon containers of the White River treatment plant effluent were stored at 68°F and 80°F. Identical containers were used for both waters with a free chlorine residual and a chloramine or combined residual. Figure 1 illustrates the persistence of chlorine residuals in relation to both time and form. Chlorine residuals were determined by starch-iodide titration as specified by *Standard Methods*. As you can readily see, the water temperature and the form of residual make a substantial difference in the persistence of the residual. This work was done under fairly ideal laboratory conditions. I doubt that chlorine residuals in our distribution piping would persist as well.

For comparative purposes, we ran a similar test on our best well water supply. Figure 2 illustrates the difference in the persistence of free chlorine residuals between our surface water treatment plant effluent and this particular well water supply. These findings correlate perfectly with the work done in Detroit by Mike Taras on the chlorination of nitrogenous compounds.

Another major obstacle for the maintenance of high water quality throughout the distribution system is the design criteria. Pipe sizing is designed usually to meet fire flows on top of normal system demands. This unquestionably is a deterrent to desirable displacement, particularly in a remote residential or low water use area. The location of sizeable elevated or ground storage facilities on this piping network adds still another obstacle as positive circulation is not normally achieved. In a system such as ours, which includes 15 different pressure districts, the division valving creates additional dead ends to those normally encountered in a system that is subject to substantial growth in all directions. We have a total of over 1750 dead end situations in our system. I mention these factors which adversely affect circulation within the system only to emphasize the complexity and magnitude of the problem we face.

Despite our utilization of a chloramine residual during the summer months, we still lose our residuals in distribution system storage reservoirs and in remote portions of the system. In recognition of this problem, we have installed supplemental chlorination equipment in all booster pumping stations and elevated storage tanks that have been constructed on the system since 1956. These installations have become more sophisticated as our experience and confidence have grown with unmanned automatic chlorine feeding systems. They are normally in operation from about May through November.

We do perform all of our own bacteriological analysis generally as prescribed in *Standard Methods*. The laboratory personnel, equipment, and procedures are all inspected and approved annually by the Indiana State Board of Health. Following an extensive study conducted in our laboratory in the mid 1940's we selected Lauryl Tryptose Broth for all sample tubes on the basis that it resulted in less false presumptives than Lactose Broth on our particular waters. We do make practice of carrying to completion the testing of gas formers from finished water samples. For over ten years we have

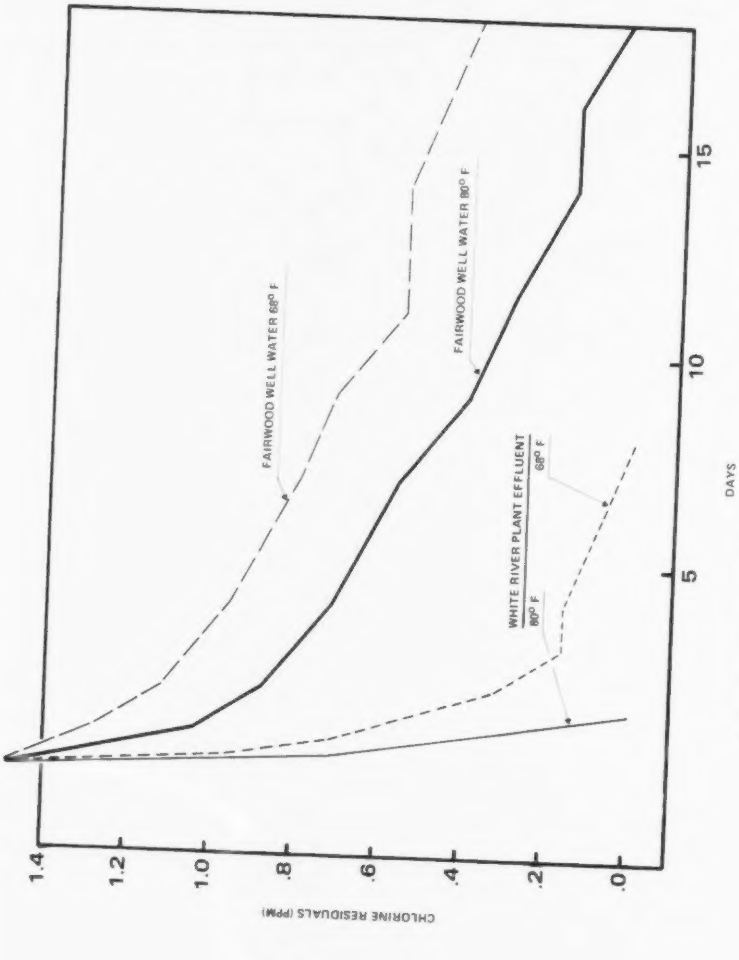

Figure 2. PERSISTENCE OF FREE CHLORINE RESIDUALS AFTER 5 HOUR CONTACT TIME.

run parallel comparisons between the membrane filter technique and multiple tube for various selective locations. As a result of this study we have elected to stay with the multiple tube procedure for all of our bacteriological analyses. We believe it is the more sensitive for our particular situation. This should become apparent as we examine the data which will follow.

Certainly one consideration in the bacterial quality of the finished water is the bacterial quality of the source. Since the White River provides approximately two-thirds of our total production, let's examine this supply from a bacteriological standpoint. Table 1 enumerates the monthly average MPN for the 10-year period 1967-76. From the table it is fairly obvious that the quality is subject to considerable variation. Generally the months with the greater numbers reflect a period of higher surface runoff. The last two months of 1976 correspond to an extremely dry period with neglible surface runoff. By some standards this would not be considered a particularly desirable source. However, it happens to be the major waterway through the central portion of Indiana.

In recent years we have also been testing our raw waters for the more specific type fecal coliform organisms. Table 2 illustrates the results of this particular series based on twice a week sampling for the past year. Monthly average numerical comparisons are shown for the MPN as determined by the multiple tube procedure, the MPN as determined by the membrane filter technique utilizing M-Endo Broth, the fecal coliform count utilizing M-FC Broth and incubation at 44.5°C, and fecal strep. counts utilizing both M-Enterococcus and KF media. As you can readily see the fecal coliform numbers are significantly below those obtained for the total coliform. For the membrane filter, fecal coliform, and fecal strep. determinations between .5 and 5 ml of raw water are used depending upon raw water turbidity and bacteriological quality.

Table 3 illustrates the bacteriological results of the treatment plant effluent and distribution system samples over a 50 year period at 5 year intervals. WR indicates the effluent samples from the White River treatment plant, and FC indicates the effluent samples from the Fall Creek treatment plant. The Fall Creek rapid sand treatment plant was placed in service as a secondary supply in 1941. At the White River plant the initial rapid sand filters were placed in service in 1926. Prior to that all filtration was through slow sand filters since about 1905. Additional rapid sand filter expansion at this plant occurred in 1953, 1959, and 1972. Back in 1925 and 1930 the chlorine residuals leaving the plant were in the range of .10 ppm. This would be considered an extremely low range according to our current standards. I would sincerely doubt that it was a breakpointed or free chlorine residual. A good understanding of this chlorination phenomena did not occur until the mid to late 1930's. In reviewing this data I feel it is apparent the conventional treatment process with responsible operators can provide an exceptionally safe bacteriological product regardless of variations in the quality of the source. Likewise the data indicates that this quality does tend to deteriorate slightly in the distribution system. While we are able to show a significant and consistent improvement in the coliform recovery from the system samples, we have not shown the same degree of success with regard to the total (standard) plate count. Undoubtedly the improvement in coliform recovery is largely the result of leaving the treatment plants with higher levels of chlorine residuals over the years.

TABLE 1. MOST PROBABLE NUMBER OF COLIFORM PER 100 ML RAW WATER — WHITE RIVER (1967-76).

	1967	1968	1969	1970	1971	1972	1973	1974	1975	1976	Average
January	33,236	36,553	278,942	15,538	15,574	54,520	59,423	66,900	49,231	51,400	66,132
February	47,513	45,342	65,592	51,654	169,065	9,300	12,363	23,179	38,383	218,373	68,076
March	31,672	6,648	5,984	1,570	9,876	90,910	43,746	10,666	17,504	25,222	24,380
April	25,616	45,921	23,908	15,203	1,174	40,391	11,321	33,456	23,488	1,108	21,153
May	18,050	14,970	3,299	3,566	1,511	6,608	2,958	9,330	6,415	685	6,739
June	2,196	4,909	4,226	5,245	3,327	7,209	9,819	9,732	3,151	3,860	5,367
July	1,279	1,989	8,852	6,962	24,067	9,104	5,464	6,478	5,709	3,611	7,352
August	654	6,174	4,842	6,796	5,747	6,343	16,366	7,222	7,010	2,236	6,339
September	673	1,134	6,854	15,557	11,041	10,663	5,006	6,854	2,451	2,450	6,268
October	1,888	4,100	13,311	11,588	4,092	6,561	4,611	7,677	2,579	1,952	5,836
November	1,565	14,799	131,983	7,674	2,862	143,358	40,124	13,832	5,018	677	36,189
December	202,394	64,300	16,265	11,469	70,150	217,664	225,229	86,498	57,085	564	95,162
Average	30,085	20,378	46,564	12,581	25,671	43,545	35,587	23,590	18,312	25,402	28,172

TABLE 2. RAW WATER—WHITE RIVER (1976).

	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.	Avg.
Fecal Coliforms	41,977	262,363	40,744	2,948	683	3,481	5,377	2,454	13,354	4,464	629	660	31,881
Fecal Streptococci	939	4,419	2,788	617	5,328	3,833	1,407	4,400	1,992	2,837	578	189	2,890
1. M-Entero-	2,502	1,463	1,450	225	290	724	430	217	192	252	118	63	691
coccus Agar	874	744	367	75	195	500	378	494	429	633	101	149	423
2. KF Agar	1,287	840	693	200	219	541	441	329	300	367	301	389	504
Turbidity (RW)	25	90	80	45	45	50	75	55	45	30	5	5	45

TABLE 3. ANNUAL AVERAGES.

	35° Plate Count Plant Effluent		% Coliform Plant Effluent		35° Plate Count Distribution System	% Coliform System Distribution
	WR	FC	WR	FC		
1975	1	3	.06	.28	75	.14
1970	2	1	.01	.01	300	.29
1965	2	2	.02	.17	18	.29
1960	2	2	.01	.00	100	.32
1955	1	1	.01	.01	300	.44
1950	2	1	.05	.03	110	.67
1945	2	2	.18	.03	60	.80
1940	3		.36		85	1.48
1935	5		.00		11	1.40
1930	4		.00		210	1.50
1925	10		.00		325	2.32
AVG.	3	2	.07	.08	140	.88

Table 4 examines the average monthly distribution of system bacteriological samples for the 25-year period (1951-1976). We examine approximately 375 samples or 1875 tubes per month from the distribution system. This is 125 samples over that required by the Drinking Water Standards on the basis of population served. The numbers under the coliform heading represent the average number of tubes that completed as coliforms for that particular month. Naturally you recall that a total plate count limitation was proposed in the Interim Primary Drinking Water Standard. We objected rather strenuously on the basis that our historical record indicated we regularly recovered counts in excess of this number, and we were unaware of any health related significance. The second row of numbers indicates the monthly average total plate counts. With a few individual samples reaching 10,000 colonies it is not difficult to reach a high monthly average. The lower row of numbers indicates the number of plates or samples which would have exceeded for the month the proposed 500 plate count. Quite obviously the deterioration in bacteriological quality is a seasonal one for us peaking during the hot summer months. Water temperature plays a part in a number of influential parameters.

Table 5 provides an analysis of all distribution system samples on a monthly basis for the past ten years (1967-76). The seasonal trend which appeared so traditionally in the previous slide is not so apparent in this one. However, we can determine that during this particular period that gas formers were recovered in .29% of the tubes examined. Likewise that the number of coliforms which completed averaged .19% of the tubes examined or almost exactly two-thirds of the gas formers recovered. The irregular pattern shown in this slide perhaps introduces still another variable. Certainly the mishandling of the sample in either collection or preparation can lead to entirely erroneous results which nevertheless influence the quantitative measure of water quality or safety. While this does have a slight impact on our monthly averages, it might be critical for a small utility with a limited number of samples.

Table 6 analyzes each sample which resulted in gas formation in three or more 10 ml portions for the years 1970-76. For each sample is shown the chlorine residual, total place count, number of tubes showing gas formation, and the number of tubes completing as coliforms. Almost identical data is

TABLE 4. 25-YEAR MONTHLY AVERAGE (1951-76) DISTRIBUTION SYSTEM SAMPLES.

	Jan.	Feb.	Mar	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
COLIFORMS	3	4	3	4	6	9	10	12	9	7	3	3
TOTAL PLATE COUNT	8	8	6	6	41	78	318	439	262	133	52	18
NO. PLATES EXCESS 500	1	1	0	1	5	10	25	38	31	19	7	6

TABLE 5. DISTRIBUTION SYSTEM SAMPLE ANALYSIS (1967-1976).

	No. SAMPLES	No. TUBES	GAS FORM	No. of Samples						TOTAL COLI
				0/5	1/5	2/5	3/5	4/5	5/5	
Jan.	3,782	18,910	26	3,764	13	5	0	0	0	23
Feb.	3,596	17,980	44	3,575	11	4	3	1	2	42
Mar.	3,841	19,205	22	3,831	7	1	0	0	2	19
Apr.	3,690	18,450	53	3,667	13	3	3	1	3	47
May	3,779	18,895	64	3,756	12	3	2	2	4	52
June	3,769	18,845	27	3,760	7	0	0	0	2	17
July	3,765	18,825	98	3,733	21	5	3	2	1	53
Aug.	3,882	19,410	77	3,853	17	5	1	4	1	51
Sept.	3,623	18,115	110	3,596	17	3	3	4	0	48
Oct.	3,961	19,805	53	3,950	8	2	0	0	1	17
Nov.	3,440	17,200	40	3,423	13	2	0	2	0	25
Dec.	3,698	18,490	46	3,667	28	2	0	0	1	37
Total	44,826	224,130	660	44,575	167	35	15	16	17	431
Avg.	3,736	18,680	55 .29%	3,715	14	3	1.3	1.3	1.4	36 .19%

TABLE 6. DISTRIBUTION SYSTEM COLIFORM RECOVERY (3 OR MORE—10 ML) ANALYSIS.

Date	Sample No.	Cl_2 Res.	SAMPLE			CHECK SAMPLE		
			TPC [1]	GF [2]	COLI	TPC [1]	GF [2]	COLI
5-20-76	52	.80	35	5	5	0	0	0
8-18-76	140	.36	11	3	3	1,200	0	0
9- 9-76	146	.12	5,400	4	4	85	0	0
4-25-75	151	1.09	1	3	3	2	0	0
5- 1-75	139	.92	110	5	5	1	0	0
9- 2-75	302	.76	1	3	3	0	0	0
5- 9-74	139	.88	240	5	5	1	1	1
5-31-75	140	.40	70	4	3	1	0	0
9-23-74	302	.76	1	5	4	1	0	0
11-18-74	302	.72	1	4	4	1	0	0
12-11-74	42	1.00	115	5	5	1	0	0
5- 9-73	33	1.04	2	4	4	1	0	0
2-22-72	52	1.04	6	3	3	5	0	0
2-24-72	139	.80	19	5	5	5	0	0
4-17-72	139	.64	650	5	5	1	0	0
7-17-72	148	1.08	1	3	3	1	0	0
4-29-71	145	.28	1	5	5	1	0	0
4-30-71	43	.40	10	5	5	1	0	0
6-15-71	302	.64	1	5	5	1	0	0
7-12-71	58	.08	5,400	4	4	11,000	0	0
8-31-71	56	.04	55	3	3	600	0	0
2- 9-70	148	.76	1	5	5	1	0	0
5-11-70	302	.24	1	3	3	1	0	0
5-15-70	61	.20	55	5	3	1	0	0
6-19-70	150	.72	55	5	5	18	2	1
7- 8-70	150	.84	4	3	3	N.A.	0	0
8-21-70	146	0.00	14,000	4	4	N.A.	0	0
8-25-70	62	.12	6	4	4	45	0	0
11-30-70	146	.40	1	4	4	1	0	0

1. Total Plate Count
2. Gas Formation

TABLE 7. DEAD END SAMPLE COMPARISON WITH TIME.

Date	No.	Cl$_2$ Res.	IWC LAB (2 HR.)			EPA LABORATORY			
			TPC[1]	GF[2]	COLI	TPC	GF	COLI	MF
6-24-75	1	0.00	4,900	0	0	110,000	5	3	0
	2	.54	1	0	0	540	0	0	0
	3	.20	0	0	0	730	0	0	0
	4	.12	2	0	0	650	3	2	0
7-16-75	5	.12	0	0	0	240	0	0	0
	6	.12	1	0	0	480	0	0	0
	7	0.00	44	0	0	21,000	2	2	0
	8	0.00	300	1	1	760	0	0	0
	9	.12	300	0	0	30,000	0	0	0

1. Total Plate Count
2. Gas Formation

TABLE 8. WHITE RIVER SUPPLY—1975.

	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
Temp-F	38	38	43	51	66	73	77	79	68	58	52	40
Turb.-Raw	46	90	55	85	90	105	90	70	45	35	25	30
MPN-Raw	49,231	38,383	17,504	23,488	6,415	3,151	5,709	7,010	2,451	2,579	5,018	57,085
Algae-Raw	1,252	934	1,485	5,779	13,167	7,632	10,953	12,431	15,503	7,358	3,982	3,263
Alum-#/MG	224	260	133	193	215	182	249	403	185	116	114	172
Chlor-#/MG	50	43	44	46	60	64	81	94	72	60	46	59
Turb.-Sett	1.5	1.6	1.9	1.5	1.4	1.7	1.9	2.1	1.9	1.5	1.6	2.2
Algae-Sett	212	139	62	450	1,232	551	1,497	1,744	1,463	1,274	272	328
Turb.-PE	.36	.35	.23	.26	.34	.25	.57	.85	.37	.30	.26	.35
Algae-PE	25	14	9	25	94	54	165	158	99	90	23	26
Coli-D.S.	4	1	0	7	9	0	0	1	4	0	0	5
TPC-D.S.	12	9	3	11	28	3	180	450	90	31	65	4
TPC>500	1	2	0	3	4	0	13	39	16	7	4	0

also shown on each check sample. As you examine this information, it becomes apparent that regardless of your theory or beliefs there exists some exceptions. My own interpretation is that a majority of these unsatisfactory results are due to mishandling either during sample collection or sample preparation. This impression primarily centers around the relatively high chlorine residuals occurring in some of the samples at the time of collection, and the conviction that this range of residual is not conducive to significant bacterial growths. Likewise the generally satisfactory results recovered in the check samples.

During the summer of 1975 an EPA, Cincinnati Laboratory team was at our utility making some virus studies. The bacteriological laboratory back in Cincinnati requested that we collect several dead end water samples, and return them to Cincinnati with the virus team. Two sets of samples were collected from nine separate locations. Each sample was collected from the nozzle flow from a dry barrel fire hydrant as soon as it was anticipated that the branch line had been displaced. This is obviously an undesirable sample point from a sanitary standpoint. Sample preparation in our lab was within two hours. I have no definite knowledge of the time interval prior to preparation in the EPA lab, but it could have been close to 24 hours after collection. Table 7 illustrates the results from both labs with the primary variable being time. This is still another obstacle which a small utility faces in getting a water sample to a central laboratory for analysis, and according to some authorities presents a major handicap in obtaining representative quality results.

Table 8 is a brief monthly summary looking at a number of raw water parameters which may have an effect on finished water and distribution system water quality. The year of 1975 was not a particularly bad year relative to water quality, but it does illustrate one previously unmentioned parameter which reportedly has a significant impact upon bacteriological quality. The parameter that I would like to focus on is that of turbidity. Turbidity is one of the most nebulous for quantitative comparison. Turbidity comes in all sizes, shapes, and charges. Although cold water temperatures do interfere with coagulation, silt particles accompanying soil runoff are normally relatively easy to remove. However the smaller the particle size, the more difficult it is to remove. This is demonstrated by comparing the algae counts for raw, settled, and filter effluents. For our supply the most normally predominant organisms are diatoms such as *Cyclotella, Synedra,* and *Navicula.* As you can see from the data presented we fairly consistently remove 88% of the algae by coagulation and settling. The data also indicates that 92% of the remainder are removed by filtration. Raw water algae counts as high as 50,000 to 100,000 organisms per ml have been recorded in the past. Our record over several years on raw, settled, and filter effluent algae samples, indicates the percent removal statistics hold fairly uniform. Naturally, the greater the raw water concentration the higher the concentration will be in the finished water resulting in higher turbidity readings. If you will look closely at the month of August, you will see it was the month with the highest effluent turbidity even with a twice average alum dosage for about an average raw water turbidity. In the statement of Basis and Purpose for the Proposed National Interim Primary Drinking Water Standards it states "A properly operated water treatment plant employing coagulants and granular filtration should have no difficulty in consistently producing a finished water conforming to this limit." We are one utility that can meet the limitation of one (1) Turbidity Unit on an annual basis, but from our historical data we know we will exceed that limitation on a monthly basis under certain raw water quality conditions.

It has not been my intention to confuse the issue by the presentation of limited operating data pertinent to the subject of bacteriological quality. Rather that the subject is a complex one requiring a knowledgeable interpretation of perhaps misleading results. The Indianapolic Water Company historically has met and expects to continue to meet the proposed coliform standard in the Primary Drinking Water Regulations. Please remember that PL 93-523 is known as the "Safe Drinking Water Act" and not the "Pure Drinking Water Act."

The most distasteful portion of the Act to me and many other water utility operators is that of public notification. No responsible operator would hesitate to forewarn their consumers when a known hazard exists, but to do so based upon an erroneous test does nothing but discredit the utility or the industry. It is my personal observation that public notification should be made only after a careful review of data indicating an MCL has been exceeded by a knowledgeable public health authority, and that a hazard does or did exist.

Another area of the Act that I question is the responsibility of the utility to maintain water quality to the tap. This really is beyond the limits of any control by the utility, and actually falls on private property. The water utility does not establish plumbing codes, approve building plans, or perform building inspections during or following construction. Likewise it has absolutely no control over the remodeling or repair of existing plumbing within the consumers property. We know from first hand experience that these actions can result in unsatisfactory and unrepresentative bacteriological quality.

From the standpoint of elapsed time and our operating data the second daily check sample would appear superfluous. In the event of plumbing repair contamination introduced from within the sample structure, samples collected from adjacent sites may prove to be much more representative of actual water quality in the system. If a bad sample is recovered in a small system late in the month, there is no reasonable way an operator can collect enough samples to improve his monthly average. If the bad sample occurs early in the month, he may have a chance. If there is a suspected cause of sample contamination in the original sample, it makes more sense to me to disregard it in the record and retain the results of a satisfactory check sample. Rather than retain the results of an unrepresentative sample, and not even consider the results of the check sample. I realize this all boils down to the numbers game in trying to meet a hard and fast regulation. I think it would be well to point out that most responsible water utilities operate according to well documented and proven practices and principles evaluated to fit their own particular situation. As long as all these systems perform properly, satisfactory bacteriological results will generally follow. However, if a fault occurs in the system and unsatisfactory bacteriological quality results, no number of additional samples will correct the fault.

I sincerely appreciate the opportunity to express my obviously prejudiced views on this subject to this group today. I am equally grateful that it is not my responsibility to establish bacteriological quality standards to protect the public's health under all the known variable conditions that exist throughout the nation.

Compliance with PL 93-523 Coliform Standards

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Many people have taken the coliform standards for granted. The advent of PL 93-523 and its mandatory compliance requirements have now begun to raise questions in the minds of some about problems of attainability and compliance with these standards. This presentation will briefly review the development of certain aspects of the coliform standards and discuss the program of compliance in California.

In 1925 Treasury Department standards for drinking water were revised and the coliform standards as we know them were first presented. The membership of the advisory committee reads like a Who's Who of Sanitary Engineering including: George C. Whipple, C. A. Emerson, George W. Fuller, Charles G. Hyde, Edwin O. Jordan, Harry E. Jordan, Milton J. Rosenau, Robert S. Weston, C.-E. A. Winslow, and Abel Wolman—a most impressive group.

The report of this committee is brief and exceedinly interesting, and is presented in full as follows:

> "The task referred to this committee by the Surgeon General of the Public Health Service is to formulate definite specifications which may be used by the Public Health Service in the administrative action which it is required to take upon the supplies of drinking water offered by common carriers for the use of passengers carried in interstate traffic. The recommendations submitted apply, therefore, only to this special case, and are not proposed for more general application.
>
> "Since the purpose of the supervision which the Public Health Service exercises over these water supplies is to safeguard the health of the public, the examinations and specific requirements herein proposed have reference chiefly to forming a judgment of safety, and are designed especially to afford protection against the most serious danger which is associated with water supplies, namely, that of infection with typhoid fever and other diseases of similar origin and transmission. Less emphasis has been placed upon physical and chemical characteristics affecting the acceptability of water with respect of appearance, taste, and odor, because these are matters of less fundamental importance and because, in actual experience, the water supplies which come under consideration, if satisfactory from the standpoint of safety, will usually be found satisfactory with respect to physical and chemical characteristics.
>
> "The first step toward the establishment of standards which will insure the safety of water supplies conforming to them is to agree upon some criterion of safety. This is necessary because "safety" in water supplies, as they are actually produced, is relative and quantitative, not absolute. Thus, to state that a water supply is "safe" does not necessarily signify that absolutely no risk is ever incurred in drinking it. What is usually meant, and all that can be asserted from any evidence at hand, is that the danger, if any, is so small that it can not be discovered by available means of observation. Nevertheless, while it is impossible to demonstrate the absolute

safety of a water supply, it is well established that the water supplies of many of our large cities are safe in the sense stated above, since the large populations using them continuously have, in recent years, suffered only a minimal incidence of typhoid fever and other potentially water-borne infections. Whether or not these water supplies have had any part whatsoever in the conveyance of such infections during the period referred to is a question that can not be answered will full certainty; but the total incidence of the diseases has been so low that even though the water supplies be charged with responsibility for the maximum share which may reasonably be suggested, the risk of infection through them is still very small compared to the ordinary hazards of everyday life.[1]

"The committee has, therefore, taken this better class of municipal water supplies as its standard of comparison with respect to safety and proposes, as a fair objective, that the water supplies furnished by common carriers to passengers in interstate traffic be of comparable safety. As regards protection of the traveling public, such a standard is fair, since it implies that the use of the water supplied to them in travel shall not add to the almost negligible risk which is ordinarily incurred at home by those who habitually use water supplies of somewhat better than average quality. From the standpoint of the carriers also, this standard is believed to be fair and reasonable, since it refers to water supplies which are actually obtainable in all sections of the country and from a great variety of sources.

"The next and principal task of the committee has been to set up objective requirements which will conform to this general standard of safety; that is, requirements which will ordinarily be fulfilled by the municipal supplies of epidemiologically demonstrated safety which constitute the standard of comparison, but will exclude supplies of less assured safety. Since there is no single and measurable characteristic of water supplies which bears any known and constant relation to actual safety, the standard recommended is composite, including certain requirements relative to the source and protection of the water supplies in question as indicated by a careful sanitary survey, and certain other requirements relative to bacterial content as shown by standard tests.

"It is anticipated that little objection will be raised to the requirements laid down as to source and protection, at least to their general intent, because they are based upon well recognized principles of sanitary engineering, and because they are necessarily stated in general terms which imply a rather broad consideration of each supply from all angles and the exercise of discretion in forming an ultimate judgment of its fitness. The bacteriological standard, on the other hand, is stated in definite quantitative terms. This is unavoidable if such a standard be included at all, since the methods of bacteriological examinations are quantitative and yield

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[1] This evidence actually proves only that the water supplies in question have been generally "safe" in the past during the period of low prevalence of infection. The likelihood that they will continue to be equally or more safe in the future must, of course, be reckoned from other considerations, such as the probability of future change in the pollution of their watershed, the character and consistency of their protection, etc.

> results in the definite terms used in the standard. However, in view of the well-recognized principle that the significance of bacteriological examinations is variable, and must be interpreted with due regard to all other facts known about the particular water supply in question, the objection may be raised that a rigid application of this standard will arbitrarily exclude a considerable number of water supplies which conform to all other requirements and which competent opinion will consider to be quite safe. The validity of this criticism is recognized, but it is not considered of sufficient force to require or justify the lowering of the bacteriological standard proposed. This viewpoint appears proper when it is recognized that the definite terms of bacteriological quality in which this standard is expressed represent only agreement as to safety, and not as to limiting values beyond which demonstrable or even presumptive danger lies. Between the point on which the committee is in agreement as to the assured safety of water supplies and the point at which agreement could be reached as to their dangerous quality is a wide zone. Within this zone lie many water supplies which, if considered in the light of available evidence from all angles, are believed to be as safe as other supplies which conform to all the bacteriological requirements.
>
> "The committee, therefore, considers it preferable to recommend that in actual practice the bacteriological standard be applied, as are other requirements, with some latitude; in other words, that supplies which, on rigid inspection are found to be satisfactory in other respects but fail to meet the bacteriological standard, may be accepted in the discretion of the certifying authority. In view of the character of the personnel entrusted with the responsibility for investigation and administrative action, the committee feels assured this procedure is preferable to the alternative of rigid and automatic application."

In one of the appendices of the report, the committee's philosophy on attainability of the standards is presented as follows:

> "As to the reasons for specifying these particular limiting values rather than some other values, either higher or lower, it is obvious that the assignment of any definite limits of bacterial content as a criterion of the safety of water supplies of diverse origin and history must necessarily be an arbitrary procedure, because the relation which the determinable bacterial content bears to the actual safety of a water supply is variable and to some extent indeterminate. Therefore, all that may be claimed for the standards proposed is that, in the judgment of this committee, they are reasonable; that is, are consistent with the other requirements specified as to source and protection of the water supplies in question, afford an ample guarantee of safety, and can be met without too costly and burdensome effort. In connection with this last consideration, the committee has analyzed the records of daily examinations of a considerable number of municipal water supplies for the years 1919 to 1922 and finds that a substantial majority of them conform to both requirements of the standard."

Whether or not these statements reflected general professional opinion on the subject or whether they molded professional opinion cannot be said, but most certainly as I saw the standards applied in the decade of the 30's and on, practices in application of the standards were consistent with these views.

The drinking water standards, from the very beginning, were adopted under the authority of the Interstate Quarantine Regulations and were the standards for the water to be used on common carriers, railroads, vessels, etc., carrying passengers in interstate traffic. In addition, these coliform standards (of course, the organisms were called *B. coli* in the 20's and early 30's) were generally recognized as the nation's drinking water standards and applied by the states. I do not know the details of the history of the use of these standards state-by-state. I believe that gradually more states formally adopted these standards, as their own, by reference. As to compliance and enforcement, undoubtedly this was a mixed bag around the country. First, speaking just about compliance for waters used on interstate carriers: Until 1942, sampling for compliance with the standards was done partly at the source and partly in distribution systems. Unquestionably, practices varied from place to place. It is likely that, as with places studied for the 1925 standards, most of the sampling was done at the point of production of the water, at treatment facilities or at ground water sources. It is significant that as to compliance, water systems will more readily comply if sampling is done at sources than if sampling is done in distribution systems.

The 1925 standards contain no directions or limitations with relation to sampling location. It is likely that the practices at the time were to sample at the point of water production and to a lesser degree in distribution sysems. The 1942 standards, in recognition of the Chicago amoebic dysentery outbreak attributed to cross connections, required that the bacteriological examination of water be of samples collected throughout the distribution system. In other words, source samples could not be utilized in considering compliance. Thus, even though the coliform numbers for the standards were unchanged from the 1925 standards, in fact these standards were more difficult to attain.

At this time compliance was based on annual results—a summary of all samples collected during the reporting year. Sometime in the late 1940's the Public Health Service administratively changed the ground rules and thereupon required compliance on a month-by-month basis. This also increased the difficulty of compliance.

The remainder of this discussion concerns the conduct of the coliform monitoring-compliance program in California through the years. The Bureau of Sanitary Engineering was created in 1915. At that time little water was receiving treatment. The city of Sacramento obtained water without treatment from the polluted Sacramento River. According to the July 1915 monthly bulletin of the California State Board of Health, Sacramento had a death rate from typhoid averaging about 53½ per 100,000 annually for each five year period since 1895 although some of these were "imported" cases. The case rate, as contrasted to the death rate, was about ten times as high. The typhoid death rate for the entire State was 13.6 per 100,000 population. The program emphasis for the Bureau of Sanitary Engineering for the next 15 years was to get sewage out of water supply sources, to get adequate treatment for surface water sources, and to eliminate hazardous wells. The first statewide comprehensive sanitary engineering survey of community water systems was made in 1930. Beginning in the mid-30's with the advent of WPA and PWA projects, program emphasis was transferred to development of sewage treatment facilities to replace raw sewage discharges. When I came to work in the State Health Department in 1938, the water supply program was focused on (1) dealing with "problem" systems to get correction of major defects, and (2) annual inspections of the interstate carrier systems. I estimate that at that time the Department had probably four or five hundred systems under surveillance.

Efforts were made to bring systems into compliance with the coliform standards when they were not met, but the major emphasis was on improving physical works and operating procedures that were likely to result in, or had the potential for, production of poor quality water. Compliance with the bacteriological standards was (and still is) a secondary matter. Public health is founded on the solid rock of prevention. Our philosophy is that the safety of public water supply is assured through attention to the development of satisfactory facilities and existence of capable management and operator personnel. Sampling and analysis is only one part of the surveillance process and provides after-the-fact information, analogous to watching the scenery from the rear observation platform of a train. In 1938 a few utilities—the larger muncipally owned ones, such as Sacramento, San Francisco, East Bay MUD, Los Angeles, and San Diego—and 2 or 3 major investor-owned utilities such as California Water Service, carried out routine bacteriological monitoring programs with samples analyzed in their own laboratories. For the rest of the systems, the State Health Department did infrequent sampling. Samples were collected at least once a year at the interstate carrier systems, which numbered 100 or so. For the rest, water systems were visited at least on a decennial basis, and more often where the problems were known to exist. At the time of the decennial visit, a thorough investigation was made of the physical works, facilities operation, and relevant operating records to the extent that any existed. Bacteriological and chemical sampling also was done. The water at some of the places met the coliform standards, and at other places, it did not.

As the years went on, and additional manpower became available—and this was always a hard struggle—the program was tightened up. Gradually a policy evolved of requiring all water utilities to collect water samples in compliance with the Drinking Water Standards frequency requirements, and to have those samples analyzed in "approved" laboratories. Beginning in the early 60's, we initiated an aggressive program of requiring compliance with the frequency standards. As more places complied, we began collecting and organizing statewide statistics with relation to compliance with the bacteriological standards. As of about 1968 approximately 30% of the water systems under our jurisdiction—1100 with 200 or more service connections—failed to meet the coliform standards one or more months out of the year. Perhaps a third of those not in compliance failed multiple months, some failing as much as 6 or more months of the year. For the past 8 years, we have worked very aggressively to get compliance, following up month-by-month with those places which failed to meet the coliform standards the previous month. Now, after about 8 years of quite energetic effort, though with no efforts at legal action, the statistics for 1976 show about 8.9% (97) of approximately 1100 systems failing to meet the coliform standards one or more months. Last year 2 places failed to meet the standards for 5 months; 2 failed for 3 months; 14 failed 2 months; the remainder (79) for only 1 month. These statistics are low since compliance has been calculated from running series of the last 20 samples.

As I have indicated, the "state" program deals with water utilities with 200 or more service connections. Local health departments deal with the less than 200 service connection systems, and the current inventory records about 4400 community water systems under local health department jurisdiction. These systems are only partly under routine bacteriological surveillance, and statistics are available for calendar 1976 for 2136 of these systems. The results show that 32 systems failed the coliform standards 6 or more months; 209 systems failed 2-5 months; and 162 systems failed 1 month. In summary, 403 systems failed 1 or more months, amounting to approximately 981 system/month failures. This would have produced that many public notifications of failure to meet coliform standards. As noted above, these statistics are on the low side because compliance was calculated from running series of the last 20 samples. In addition, it is likely that these statistics in part represent the better small water systems. If all systems were under bacteriological surveillance, the proportion of failure to comply would be higher. Most certainly there is a big problem of bringing these systems into compliance with the standards.

A final comment—and an important one—what are the causes for failure? Of course, they are varied. Almost no failures occur at the point of production at water filtration facilities. Some failures are traced to malfunctioning of chlorination facilities at small places with surface water sources and no filtration facilities. A few failures are traced to coliform "contamination" of wells. (We will not accept chlorination of coliform-producing well water as a method of compliance. Wells must produce coliform-free water or be cut off the system.) Other causes for "high" coliform are storage facility defects; for example, birds gaining entry, work on distribution lines (main breakers, etc.)—coupled with unsuccessful main disinfection—and, finally, the elusive "sampling error."

Follow-up samples are required at all places failing to meet the standards. In a substantial fraction of places, the follow-up samples are free from coliform, and the original positives are attributed to "sampling error"—or to some transitory condition of water quality. If the cause of the positive sample is a real thing, further positives may result—though sometimes sporadically. As to cross connections producing coliform contamination, few if any cross connections have been discovered through bacteriological sampling. This is not to say that cross connections have not resulted in contamination of water systems—the reverse is our experience.

Since 1970 California statutes have contained a public notification section as follows:

> "When it is determined by the department that it is in the public interest, the department shall notify a person, who has not met the standards of the department for water quality of such noncompliance, and shall require such person to notify in writing each of his customers of the department's determination that the quality of the water fails to comply with the standards, requirements, or conditions established by the department and to include any comments of the department regarding the possible dangers because of such noncompliance. The content of such statement shall be approved by the department prior to distribution. Notification by such person shall be repeated at intervals as required by the department or until the department determines that there is compliance with the standards or requirements of the department."

In 1973 the Legislature added the following with relation to public notification:

> "Upon determination by any person who furnishes or supplies water to a user for domestic purposes that a significant rise in the bacterial count of water has occurred in water which is being supplied to a user for domestic purposes within the State of California, the person who furnishes or supplies such water shall provide, at

> such person's expense, an analysis of such water within 24 hours to the county health officer and the State Department of Health. As used in this subdivision 'significant rise in the bacterial count of water' means such rise in the bacterial count of water as the department determines, by regulation, represents an immediate danger to the health of the water users."

Regulations adopted pursuant to this section define significant rise as follows:

> "(a) Significant rise in bacterial count representing an immediate danger to the health of water users is defined as an increase in coliform bacteria, when associated with a suspected waterborne illness or disruption of physical works or operating procedures.
>
> "(b) The following criteria shall be used as an indication of a significant rise in bacterial count, and is applicable to routine bacteriological samples . . .:
>
>> "(1) Whenever all five portions of more than one sample collected during a week show the presence of coliform organisms; or
>>
>> "(2) Whenever 50 percent or more of the portions of a group of two or more samples collected during a day show the presence of coliform organism; or
>>
>> "(3) Whenever the daily samples, collected following unsatisfactory samples . . ., show the presence of coliform organisms in three or more portions in each sample if more than one sample is taken, or in all portions if only a single sample is taken."

Upon a finding of significant rise, notification of both the Department of Health and the water users is required, as follows:

> "(a) When the coliform limits specified above are reached or exceeded, the water utility shall immediately notify the department by telephone of the bacteriological findings and, at the same time, shall furnish information on the current status of physical works and operating procedures which may have caused the elevated bacteriological findings, or any information on community illness suspected of being waterborne.
>
> "(b) Upon receipt of information specified in (a), the department, taking cognizance of all available information shall determine whether or not continued delivery of water would constitute an imminent danger to the health of the users.
>
>> "(1) Upon finding an imminent danger, the department shall immediately notify the water utility of this finding and of the necessity to implement the emergency notification plans . . ."

These sections have recently been modified where necessary to bring California statutes into compliance with PL 93-523.

In conclusion, an opinion on the attainability of the current bacteriological standards: These standards are stringent—only very minor defects can produce failures to meet the standards—but they are attainable when everything is in good order. Unless there is universal chlorination with chlorine residuals carried to the end of all distribution systems, a fraction of water systems will always fail to meet the standards. I consider that this eventuality does not represent a public health problem of any significance. The problem with compliance is that with current waterworks practices, a great deal of regulatory effort is necessary in order to get a reasonable degree of compliance. Meanwhile, I am sure some states have a long way to go.

Canadian Drinking Water Standards

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The desirability of having uniform drinking water standards throughout Canada has long been recognized. In early attempts to achieve this, the considerable amount of data and expertise available in the United States was utilized. This was done mainly through cooperation between national agencies and by consultation with many scientists active in the relevant fields. Our present attempt to coordinate U.S. and Canadian efforts follows this tradition.

We do not claim to represent the opinions of every agency involved in the regulation and enforcement of drinking water standards in Canada. However, as members of a major committee (formed by the Canadian Public Health Association) involved in the revision of Canadian Drinking Water Standards, we hope to present a Canadian consensus arrived at by this committee in its presentation to the Department of National Health and Welfare, Canada.

The history of drinking water standards in Canada dates back to 1923, when the Federal Cabinet passed an order-in-council which provided a standard of bacteriological quality applicable to water used for drinking and culinary purposes in vessels using Canadian inland waters including the Great Lakes. In 1930, these regulations were extended to all types of common carriers crossing international borders into Canada and inter-provincial borders within Canada. Canadian coastal shipping was included in 1937.

Drinking water standards became more generally applicable when the Department of National Health and Welfare, with the approval of the Dominion Council of Health, began to apply the 1943 United States Public Health Service Standards, pending an examination of the situation in Canada. In the interim, Canadian authorities referred to the U.S. standards of 1943 and 1962 as well as those published by the World Health Organization in

1958 and 1963. The Canadian Public Health Association (CPHA) also recognized the need for standards suitable for Canadian conditions which are not always the same as those in the United States and other parts of the developed world. It formed a Drinking Water Committee in 1966 to develop appropriate guidelines. A year later, the Department of National Health and Welfare convened an Advisory Committee on Public Health Engineering. The task of drawing up drinking water standards was delegated to a subcommittee which decided to combine forces with the earlier group formed by CPHA. With the able assistance of Mr. S. K. Krishnaswami, who was appointed principal investigator, the joint committee prepared preliminary reports and drafts for desirable standards.

Three levels of drinking water quality were designated on the basis of the premise that water for domestic use should be free from pathogenic organisms and their indicators as well as from other deleterious substances besides being palatable and free from objectionable odours, tastes and colours. The poorest of these quality levels was called "maximum permissible", the next "acceptable" and the most desirable level, which should be the goal, was called "objective". It was emphasized that the poorest quality should not be permitted for any length of time. On the basis of the detailed recommendations of the committee and taking into account other national and international standards, the 1968 Canadian Drinking Water Standards and Objectives was drawn up and published by the Department of National Health and Welfare.

The original committee emphasized the need for continuous surveillance of changes in water quality and for studies to develop basic information on Canadian conditions and requirements. In 1975, the Department of National Health and Welfare established a Federal-Provincial working group for revising the Canadian Drinking Water Standards, a task which is expected to be completed by 1978. As part of this process, a number of submissions have been made including the report by the CPHA advisory committee of which the authors are members. One of the authors (J. H.) prepared a draft for a Criterion Document entitled "Microbiological Quality of Drinking Water" based on current knowledge drawn from national and international sources. The criteria document with recommendations was finalized and submitted to the Department of National Health and Welfare.

Regulation and enforcement of water quality standards in Canada are traditionally vested in provincial agencies with the territories in the north falling under federal jurisdiction. Differences can and do exist between provinces. The situation in Ontario, for example, is complex. The Ontario Ministry of the Environment (formerly Ontario Water Resources Commission) is responsible for the overall control of regulation and enforcement of water quality in public supplies. They work in cooperation with the Ministry of Health, especially when potential health problems are encountered. Both Ministries maintain large water microbiology laboratories centrally in Toronto and smaller ones in various regions of the province. To avoid duplication of efforts, the Ministry of the Environment laboratories limit their activities to public piped systems which are under their control. The Ministry of Health laboratories deal with specimens collected by local and provincial health agencies. In addition, major municipalities such as Toronto have their own testing facilities and in these cases, the Ministries play consultative roles.

Similar jurisdictional delineations with some unavoidable overlaps exist in other parts of the country. The regulatory agencies at provincial and municipal levels follow the current Canadian Drinking Water Standards with modifications designed to suit perceived local needs and to improve the service. Some, such as the Ontario Ministry of the Environment, have developed their own objectives, which conform to the national standards but specify details of improved sample collection or additional examinations. Thus the results of the attempts to attain a measure of uniformity are encouraging.

The 1968 Canadian Drinking Water Standards specify that methods given in the current edition of "Standard Methods for the Examination of Water and Wastewater (APHA)" should be employed in the routine examination of water. It is also recommended that sampling frequencies should conform to the U.S. Public Health Services recommendations of 1962 with the proviso that frequency and location of sampling be established by the control agency or Medical Officer of Health after adequate investigation of the source, method of treatment and protection of the water supply. Raw water is considered "acceptable" only if at least 90% of the samples in any consecutive 30-day period have fecal coliform densities of less than 100 per 100 ml and a total coliform density of less than 1000 per 100 ml, although the organisms themselves may not display any such quantitative relationship.

Finished drinking water is considered "acceptable" if 95% or more of the samples tested in a 30-day period are negative for total coliform organisms when enumerated as a Most Probable Number (MPN) index by the multiple tube fermentation method. None of the positive samples should have MPN indices greater than 4 per 100 ml. Waters with MPN indices of 4 to 10 per 100 ml in more than three consecutive examinations should be immediately investigated in respect of source, treatment and other parameters. According to the 1968 standards, the membrane filter (MF) coliform test should be used to determine the potability of a given water supply only after adequate parallel testing has demonstrated that the MPN and MF methods yield equivalent information, relative to the sanitary quality of the water supply system. The MF method should be carried out on a minimum sample volume of 200 ml. Drinking water is considered "acceptable" if at least 95% of the samples in a 30-day period are negative and none of the positive samples show MF counts greater than 4 per 200 ml or 10 per 500 ml.

Any water with MF counts greater than 6 per 200 ml or 15 per 500 ml or approaching these limits in three consecutive tests should be immediately investigated. A special series of samples and the commencement of suitable and effective remedial measures must follow any deviation from MPN or MF standards. In such an event, all samples must be carried to the "completed" test procedures. In addition, the demonstration of any pathogenic organism in finished water must lead to the rejection of the supply until effective remedial treatment and disinfection have been completed. The Medical Officer of Health or the control agency may issue an order requiring boiling of water or other corrective action in appropriate situations.

There is no published information on whether water quality standards are in fact being met. However, we were able to draw upon the experience of Mr. L. Vlassoff, Chief, Microbiology Section, Ontario Ministry of the Environment. In his experience of many years involving over 500 water treatment plants, enforcement of standards has not presented serious problems. In these examinations, a simpler sampling system is used. A minimum of 8 samples per month are collected at weekly intervals from small communities but for large centres, the number of samples is at least the same as in the 1968 standards. MPN, MF or Presence-Absence tests are carried out on 100 ml samples. About 5% of the specimens require detailed testing, which includes tests for several individual bacteria. As a result of the good rapport established between microbiologists and engineers managing public water systems in Ontario, there is good cooperation in the enforcement of standards and in taking rapid remedial action as well as in carrying out additional examinations when indicated. The Drinking Water Standards specified for public supplies may not be applicable to other water supplies, which are also likely to have enforcement problems. In revising standards, this aspect should be paid greater attention.

The recommendations of the recent CPHA committee on "Microbiological Quality of Drinking water" are summarized below and the more important sections compared with the corresponding sections in the 1975 U.S. National Interim Primary Drinking Water Regulations. The detailed recommendations of the CPHA Committee are available from the Canadian Department of National Health and Welfare.*

1. Since modern technology is capable of purifying water even from heavily polluted sources, it was felt that the major concerns should be adequacy of treatment and safety of the finished water.
2. The total coliform group is preferred as an indicator of treatment adequacy and fecal coliform. measurement is preferred for monitoring raw water quality and to indicate the potential presence of pathogens at source. The present total coliform standards should be retained but a minimal sample volume of 200 ml should be stipulated. The more detailed U.S. regulations differ from this in that the recommended sample volumes are not the same and that maximum permissible contaminant levels (MCLs) are given in detail. When the new Canadian Standards are drawn up, the final recommendations are likely to show a greater similarity to the U.S. regulations.
3. The general bacterial population should be monitored regularly. The "acceptable" level should be limited to 500 organisms per ml since at higher levels, coliform measurements may not be reliable. An additional recommendation is that the definition of coliforms should include the cytochrome oxidase negative characteristic in order to exclude members of the genus *Aeromonas*.
4. A free chlorine residual of 0.3 mg per litre should be maintained throughout the system. The turbidity of water entering the distribution system should be limited to one Jackson turbidity unit. The pH should not exceed 7.5. If higher pH levels have to be maintained, it would be necessary to specify higher chlorine residuals for more alkaline conditions. When chlorine residual measurements are substituted for bacteriological examinations, they should not exceed 75% of the required examinations. In place of each coliform test omitted, three chlorine residual determinations should be carried out. It was considered desirable that the chlorine residual should be determined daily but it was left to the control agency to determine the frequency of tests. CPHA recommendations differ from the U.S. regulations in specifying a higher level of chlorine residual but this is lower than the 0.5 mg per litre required by the 1968 Canadian Standards. Another difference is in the number of chlorine residual measurements recommended for each substitution. The increase in the use of ozone as a disinfectant by water treatment plants, especially in Quebec, has to be considered in

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evaluating the merits of substituting bacteriological tests by chlorine residual monitoring.

5. While sampling frequency based on population served might be realistic and adequate for large centres, it was felt that the requirements for smaller communities are inadequate and that the minimum sampling requirement should be raised to 10 samples per month. Where this is not practical and less than 10 samples are examined, the present standards should be modified to stipulate that none of the samples should be positive for coliform organisms. When a sample is positive, corrective action should be taken immediately and tests on daily samples carried out until no coliform organisms are found in two consecutive samples. Greater emphasis should be placed on sanitary surveys to detect problems in the system. Sampling from trouble spots should receive greater attention than the collection of representative samples.
6. In view of the unacceptable degree of variation in the efficiency of different membrane filters in recovering coliforms, specifications for membrane filters are required; these should cover all the parameters that affect the recovery of these organisms.
7. Another aspect requiring attention is quality control. The use of simulated water specimens for comparative testing in different laboratories and the development of reference procedures with which methods used in the different laboratories may be compared were suggested.
8. Research into techniques for simple methods for the detection of streptococci of exclusively fecal origin has been recommended. Health hazards due to the presence of *Klebsiella* organisms in water and the possible use of fecal sterols as an absolute indicator of fecal pollution merit further study.
9. Virological examination of water was discussed at length. It was decided not to recommend virological examination as a routine procedure although the present bacteriological standards may not reflect viral contamination. It was felt that since recent technological developments are encouraging, virological studies should be carried out selectively and that virological surveys of drinking water should receive priority when developing research programmes.
10. It is necessary to determine whether standards are being met, whether bacteriological surveillance programmes meet established criteria and whether the water supply systems are adequate for providing safe drinking water. Therefore, a drinking water supply survey which encompasses different parts of Canada has been recommended.

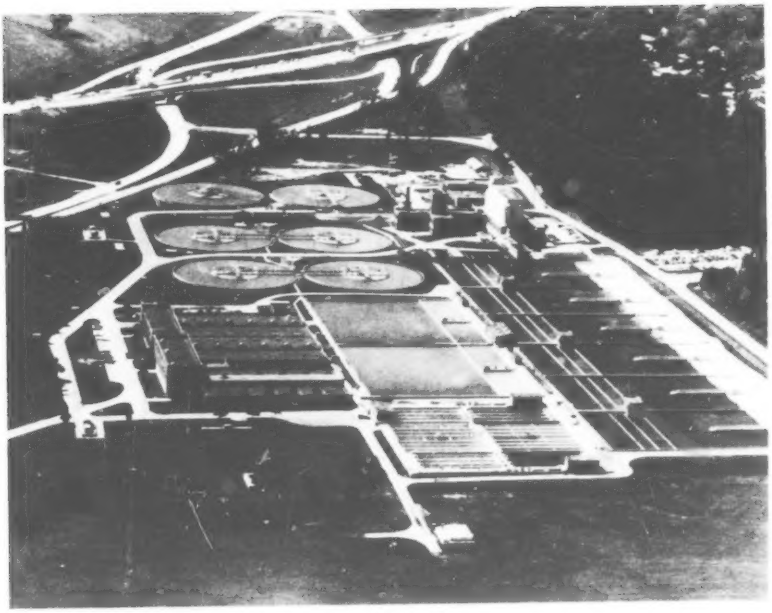
Finally, some of the differences between the Canadian position and the revised U.S. regulations reflect the fact that the revision process in the United States is at a more advanced stage. Further, the Canadian Standards provide only guidelines for regulatory agencies. When the new Canadian Standards are finalized on the basis of the CPHA recommendations and other submissions, some of these differences may disappear. Those based on differences between our situations will, however, remain. We wish to acknowledge the generous cooperation we received from Dr. P. Toft, Chief, Environmental Standards Division, Health and Welfare Canada, Ottawa and Mr. L. Vlassoff, Chief, Microbiology Section, Ontario Ministry of the Environment, Toronto.

QUESTION AND ANSWER SESSION

C.W. Hendricks, Office of Drinking Water, U.S. Environmental Protection Agency, Washington, D.C.

Do you think suppliers will sample at the rate of 8 to 10 per month?
T.P. Subrahmanyan, Enteric Virus Laboratory, Ontario Department of Health, Toronto, Canada.

The water supply utilities in Ontario, from which most of the Canadian experience is drawn, are run either by the Ontario Ministry of the Environment or by other public agencies in cooperation with the Ministry. In the case of the major municipalities, the collection of 8 to 10 samples per month has not presented significant problems. I am not in a position to comment on utilities run by private companies.



Kansas City, Missouri Water Treatment Plant

PANEL DISCUSSION

Alternative Means of Determining Compliance

Chairman: Dr. Sumner M. Morrison, Department of Microbiology, Colorado State University, Fort Collins, Colorado

Alternative Means of Determining Compliance

Sumner M. Morrison
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Microbiologists, in the area of laboratory work for the surveillance of water supplies, are sort of in the analogous position of the common expression "painting yourself into a corner". Well developed, simple, inexpensive tests for coliforms exist and are being productively applied in the control of drinking water quality for a large portion of our population. In conjunction with the work of the engineers and health officials, the laboratory work has contributed to the decline in the waterborne illness over the past 50-60 years. Now the mandate is to expand this supply of safe water to all public systems, including many, many small ones. This philosophic or political change in the need for safe water for all has created our problem. What was considered a simple, inexpensive, expeditious and effective procedure for metropolitan population centers now is considered a procedure, for many reasons already discussed, too cumbersome, costly and ineffective for the small systems. Some inconvenience, cost, dislocation and personnel training is going to occur in meeting the charge that Congress has given EPA and us as scientists and professionls with a stake in near zero risk drinking water for all.

I would like to comment about a few points that may be alternate ways of establishing compliance. Some of these ideas may be "far out" and not worthy of consideration, but this panel is to explore all possibilities.

1. While population served seems to be a very logical way to assign required tests, is there reason to consider volume treated or volume delivered for domestic use as a means of determining required numbers of tests? Systems that supply a high proportion of their water for industrial, cooling or irrigation uses are different from systems that supply domestic water only.

Similarly, is there reason to establish categories of requirements based upon type of water and type of treatment system, or perhaps the availability of engineering support? Perhaps the information we now have would not allow such an approach, but if better statistical risk data were to be accumulated we might be better able to provide the same degree of protection to all people with a more equitable cost to each.

2. The use of monthly enumeration and averaging of samples is an arbitrary use of time periods. A system using a 30 day, or even a 60 or 90 day running average would better indicate deteriorating water systems. The arbitrary calendar month will cause system operators to establish testing times to avoid getting caught with a high test in the last part of the month and other similar ways of not using good statistical sampling.

3. A major fear that seems to have been expressed is that compliance procedures do not really take into account possible sampling or laboratory error. A procedure allowing immediate retesting, perhaps for 3 consecutive days, would allow the discard of the single high coliform test without setting in motion reaction from the State and possibly public disclosure.

4. There seems to be an attempt to directly link the MCL of the bacteria to day-to-day prevention of disease. From an epidemiological standpoint it should be placed into proper context. Periodic sampling of small portions of

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water for coliforms, when coupled with proper design and operations management, has proven quite effective in preventing frequent large scale waterborne disease outbreaks. It has stood the test of time. Other alternatives to bacterial testing, as frequent chlorine residual sampling or engineering surveys only, need to be tried but in an experimental mode. Until these alternatives have stood the test of time, as has the coliform test, there should be both short-term and long-term surveillance research. To avoid again not having the answers to pertinent questions, the research must be well-designed and directed by outside as well as in-house expertise; as free as possible from self-serving bias.

5. We have heard some previous speakers analyze and criticize some of our long-established testing procedures on the basis of modern statistical analysis. Among the very important research areas that needs development in 4 above is an attempt to put such items as sampling frequency, sampling design, minimal required types of tests, correlations with human waterborne illness patterns, etc. on a solid defensible statistical basis.

6. To me, the concept of public notification when a water system appears to be in non-compliance, is a two edged sword. While it might provide appropriate pressure on the water supplier to be in continuing compliance, it could also serve to create complacency in the population of a community. In some places it could lead to severe disruption of the local community, endless litigation, suits for all types of real and imagined damage, political retribution, scare tactic competition and advertising and wasteful and unethical methods to avoid notification. In other places, with no penalties beyond notification, the local people will be successfully sold on the idea that the notices are bureaucratic garbage and they will evoke no response. In either extreme case the end result will not be safer drinking water but rising consumer costs for the legal and political maneuvers.

Far more effective would be a State program of compliance by cooperative effort with a review and emergency team to attain the compliance. By charging the costs for these emergency reaction measures back to the water supplier, the pressure would exist for staying in compliance and the chances for meeting the safe drinking water objective would be greater.

With these few remarks, I would like to introduce this afternoon's panel speakers.

The panel members for the remainder of this session are:

Dr. Warren Litsky, University of Massachusetts; Mr. Berry E. Gay, Jr., of the Division of Laboratories in the Illinois Department of Public Health; Mr. Ira Markwood, Director of Public Water Supplies for the State of Illinois; Dr. G. Wolfgang Fuhs, Director of Laboratories and Research for the New York Department of Health; and, Dr. Wesley O. Pipes, Drexel University. These individuals will present their ideas concerning alternative methods of determining compliance to the microbiology standards.

The Coliform MCL: Can We Defend It?

Warren Litsky
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During the past day and a half we have heard some excellent papers concerning evaluation of the coliform standard, detection and control of coliform bacteria and compliance with the coliform standard as they apply or pertain to the National Interim Primary Drinking Water Regulations. We have heard the pros and cons of many ageless scientific debates and we have also been privileged to the arguments of a new breed of ecologically oriented microbiologists and their rational for adopting alternative indicators of water contamination. We have listened to the advantages and disadvantages, the rational and the public health considerations of substituting the chlorine residual for the traditional microbiological procedures. After listening in awe to the parade of expert speakers I asked myself, "What will the outcome be for all this rhetoric?"

This question was not prompted by sarcasm or smugness but rather by increased activity of consumerism and consumer groups. The days are long gone when recommendations or regulations of Government Agencies were universally accepted. Unfortunately, the trend now is to distrust any government official, challenge any document put forth and above all ignore the scientists since "they are the group that got us into this present day mess." Many legal actions initiated by consumer groups against government in the name of the people have been brought to my attention with the result that a few of my colleagues are becoming extremely wealthy by acting as scientific advisors to law firms. Because of this trend and because of Section 141.32 which mandates that persons served by a community water system be notified when the system fails to comply with the applicable MCL, I predict a rash of legal entanglements, the likes of which have never been reached, and EPA better be ready for this deluge. As a mental exercise during the past few days I have asked myself, "What advice would I give to a lawyer who has been retained by a water supplier accused of exceeding the bacteriological MCL?" Realizing that the object of any court action is to prove that your opponent or the regulations, etc. that he is enforcing are incorrect, illogical, or just plain stupid, the scenario which I present may be a trifle biased, to say the least.

With these introductory remarks concluded, let me present a few items on which I would base a case were I acting in the capacity of a legal advisor.

I would most definitely attack the substitution of the chlorine residual for the coliform standard. The data in Table I demonstrate that there is most definitely fecal coliform bacteria in water showing a high chlorine residual. While these data are based on sewage effluent and combined chlorine residual, as measured by the OT test, they suggest that chlorine residual monitoring is not as infallible as its proponents wish us to believe.

Leaving this subject I would then advise the court that while we fully support the present coliform standard, we challenge the procedure by which this standard is measured! In as much as the present regulations give an unfair edge to those using the Multiple Tube Technique over the Membrane Filter Technique for enumerating the coliform bacteria, I contend that

TABLE 1. EFFECT OF CHLORINE RESIDUAL AND CONTACT TIME ON FECAL COLIFORM DENSITIES IN SEWAGE EFFLUENTS.

Plant	Effluent	Cl Residual mg/l	Contact min	F. C. MPN
A	primary	1.5	10	7900
A	primary	1.0	12	7700
A	primary	1.25	10	9200
			15	270
			20	20
A	primary	0.75	11	35,000
			15	4,900
			20	2,400
S	secondary	1.5	OF*	330
S	secondary	0.5	OF	1,100
N	primary	1.2	25	5,400
N	primary	1.1	20	16,000

* outfall

the former method will be universally employed and, in the interest of time, will limit the remainder of my discussion to the Multiple Tube Method. While the definition of the coliform group has been accepted for many years I would question whether the techniques are actually capable of isolating and enumerating this particular group. We all have heard the outcries of Dr. Victor Cabelli of EPA that members of the *Aeromonas* group, a common soil inhabitant, can not be distinguished from the coliform bacteria using the present standard methods. Thus, are we measuring coliform bacteria, *Aeromonas* species, a mixture of the two, or God knows what?

The use of the MCLs intrigues me and may also amuse the court. Case in point, Section 141.14 defines the MCL in actual numbers per 100ml when the Membrane Filter Technique is used while when the Fermentation Tube Method is used the MCL is defined as coliform present in a 10ml standard portion. In other words the MCL for the latter is defined as the number of tubes showing gas. By employing this system, the calculation of the Most Probable Number (MPN) and its very embarrassing 95 percentile confidence limits range are avoided. Whatever the reasons for this may be, the fact still remains that both the MCL and the MPN are based on positive fermentation tubes and, call it what you wish, the MCL are going to be correlated with the MPN which we have witnessed many times during the past day and a half.

Now for the MPN or MCL of coliform bacteria. As one who has been teaching sanitary bacteriology for the past twenty-five years, I have become disenchanted with the accuracy and precision of the MPN technique for coliform bacteria as outlined in *Standard Methods*. I have seen too much variation on replicate samples by technicians and students employing flawless technique. The precision of the MPN just is not there and the data in Table 2 will illustrate this. Fecal coliform determinations were made on marine waters, chlorinated sewage effluents and laboratory chlorinated sewage effluents in triplicate by a technician with ten years laboratory experience. As can be seen in Table 2, the resulting triplicate MPNs show that a ten-fold range

TABLE 2. TRIPLICATE FECAL COLIFORM MPN RANGE.

SAMPLE	MPN/100 ml		
Marine	49	310	310
	79	350	540
	94	220	350
	8	1,300	1,300
	700	1,700	7,900
Chlorinated Effluent	2,200	7,900	13,000
	49,000	130,000	490,000
	490,000	790,000	2,400,000
Lab Chlorinated Effluent	35,000	110,000	240,000
	240,000	540,000	2,400,000+
	49,000	130,000	490,000

is not uncommon. These data should not surprise anyone working with the MPN technique and yet the standard is based on this type of an analysis.

Following this I would attempt to inform the court that other investigators have published numerous papers warning of the fallacies concerning the MPN technique; but first let us consider the statistics on which the MPN was originally based.

McCrady (4) in 1915 proposed the MPN based on Bayesian statistics, a branch of statistics that modern books do not even mention. I find it impossible to explain what Mr. Bayes' theorem was and how it is involved with the MPN. However, my friends in the statistics department informed me that the following statement from McCrady's paper is the basic assumption of Bayes' statistics as applied to the MPN. I hope that you are able to make more sense out of it than I could:

> "There is a certain assumption involved in this principle which must be recognized. This assumption is that all numbers of *B. coli* are equally probable; that is, that in the long run of samples, one number of *B. coli* will appear about as often as any other number.
>
> It is to be noticed that this assumption is the same, in kind, that is involved in the determination of the 'Most Probable Number.' There, it is assumed that the 'Most Probable Number', so determined, is as probable as any other number."

In 1957, Woodward (7), then Chief of Water Supply at the Robert A. Taft Sanitarian Engineering Center, USPHS, discussed the precision of the MPN and he concluded that:

> "The lack of precision of MPN estimates of bacterial densities is generally recognized—at least by those who perform the tests. Like many other measurements used and interpreted by others than the laboratory workers who fashion them, MPN estimates are often credited with a precision they do not deserve.
>
> Precision confidence limits have been computed for three-tube and five-tube tests using multiple decimal dilutions and for the five-tube single dilution test. These limits are broader than the approximate ones based on a log normal distribution of MPN's. For a

> three-tube multiple dilution test, the 95 percent confidence limits cover approximately a 33-fold range from approximately 14 to 458 percent of the MPN estimate. For a five-tube multiple dilution test, the 95 percent confidence limits cover approximately a thirteen-fold range from approximately 24 to 324 percent of the MPN. In short, MPN estimates are not precise and many of them are inherently improbable."

Laubusch (2) in 1958 noted several limitations in the dilution technique for the determination of MPN of coliform bacteria in water:

> "Not the least of these is that the results depend on the number of tubes observed in each dilution and may be greater than the "true" coliform density (as much as 23 percent high for a series of 5-5-5 tube tests and 43 percent high for a series of 3-3-3 tests). Other limitations include deficiencies in laboratory technique and personal errors associated therewith, differences of sensitivity of alternate procedure methods, influence of one variety of organisms on another, lack of true randomness of bacterial distribution, etc. For these reasons, the MPN by the dilution method should not be treated with sacred reverence."

As to the standards Laubusch wrote:

> "The currently accepted 'Standards,' (6) which involve planting of a minimum of five 10-ml portions, were not altogether accidental. There appears to be no rational basis in the establishment of a minimum of five portions, rather than say ten or more, except that it is necessary to limit the amount of labor and materials involved to such an extent that the results are commensurate with the requisite accuracy—an entirely reasonable approach.
>
> The most accurate method of evaluating fermentation tube test results in terms of a determinate number is still open to dispute, although in more recent years this is less obvious due to the general acceptance and convenience of tables of most probable numbers from the Hoskins (1) adaption of the Reed (5) formula based on the Poisson random distribution."

Laubusch then summarizes by quoting Hoskins whose MPN tables we all have used throughout the years:

> Twenty years ago Hoskins asserted that ". . . it seems reasonable to conclude that the dilution tube method cannot result in an accurate enumeration of coli-aerogenes organisms and that, therefore, the need for an acceptable and dependable method still remains." He added further that ". . . if correspondingly as much study (as the MPN) were devoted to the development of a suitable plating phase of coli-aerogenes enumeration, it is quite reasonable to believe that this effort would yield a method superior to the present one."

Finally a short quotation from a 1975 paper by Oblinger and Koburger (5) illustrating the mathematical bias in MPN values:

> McCarthy et al. (3) demonstrated a substantial mathematical bias in MPN values relative to plate counts. With the agar plate count method as the control estimator, 10 replicate MPN's indicated an average bias of +29%, +10%, +6%, and – 4% when compared to the plate counts' arithmetic average, geometric mean, median, and harmonic mean, respectively. The precision of 10 replicate plate counts was at least three times that of replicate MPN values. Perhaps Woodward summed it up the best: "The

lack of precision of MPN estimates of bacterial densities is generally recognized—at least by those who perform these tests."

After disposing of the MPN technique I would focus on the subject of check samples. Section 141.21 (D) (2) states that when the coliform bacteria occur in three or more 10ml portions of a single sample, two consecutive daily check samples shall be collected and examined at the same sampling point. It can be argued that the check samples are required because of the belief that most positive results are due to an error in the collection process or in the laboratory analysis but seldom due to the fact that coliform bacteria were actually present in the water. I have heard speaker after speaker refer to spurious positive analysis, laboratory errors, unsatisfactory collection procedures as if they were extremely common occurrences and thus the need for check samples. I contend that the collection routine and the laboratory analysis should be such that a positive tube indicates just what it should indicate—trouble! I submit that there can be no room for error in a clinical laboratory especially during the typing and matching of blood; likewise, there should be very little room for error when analyzing finished waters. Like it or not, the check sample does not and cannot indicate the actual condition of the original sample. I see no reason why these common errors in the collection of the sample or in the laboratory analysis should be condoned as routine happenings. They need not occur and moreover they should not occur to the degree that has been indicated at this symposium.

I will be the first to admit that my arguments presented here are extremely biased and somewhat facetious. Nevertheless, these are the types of arguments that may use in future legal confrontations—and EPA had better be ready for them.

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QUESTION AND ANSWER SESSION

Mr. Kimball

What is the problem of false samples occurring from improperly sterilized bottles which are done by commercial labs?

W. Litsky, Department of Environmental Sciences, University of Massachusetts, Amherst, Massachusetts

It is assumed that employing standard methods for the analyses of water, the results obtained reflect the condition of the water. This requires that all chemicals, solutions, media and glassware be sterile. A further requirement is that the analyses be carried out by a qualified technician in order to prevent contamination of the sample, glassware, diluent or media from the environment, fingers, etc. In other words, the bacteria which are counted on the plate or by the MPN technique must originate from the sample and from nowhere else. An improperly sterilized sample bottle can in itself, be a source of bacteria thereby negating the value of the water analyses. Improperly sterilized bottles have no place in the water sampling routine. Any legitimate commercial laboratory should be able to supply sterile sampling bottles. If they can not accomplish this simple feat they should not be tolerated. They should, like the desert dwellers, fold their tents and silently slip away.

Comment by G.W. Fuhs, Division of Laboratories & Research, New York Department of Health, Albany, New York

In addition to laboratory imprecision of MPN results, environmental variability in natural surface waters (patchiness) can cause a standard deviation (in a lognormal distribution) corresponding to a factor of 4, and sometimes corresponding to one order of magnitude and more. This was determined at optimal laboratory precision (20 to 80 colonies per plate, membrane filter technique).

Participant from New Jersey

Collectors' error had been found to occur in the form of a batch of nonsterile bottles which gave false-positive results. This meant that isolated positive results could very well be in error.

G.W. Fuhs

This observation reflects a typical quality control problem and is not in conflict with Dr. Litsky's and my own views. In the case mentioned, positive results were discarded upon investigation and for a good reason. Dr. Litsky and I argue that isolated positive results should *never* be discarded *without cause*.

Alternative Means of Determining Compliance

Berry E. Gay, Jr.
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I understand there is a lawyer here in Washington, D.C. who is attempting to aid bureaucrats so they will write their agencies' regulations in plain English. With that in mind I'll make no attempt to write a regulation.

I realize that the federal government has not stipulated by regulation that all community water supplies shall be chlorinated. Illinois Environmental Protection Agency regulations do require that public water supplies shall be chlorinated. Based upon the number of public water supply samples submitted to two of the three Illinois Environmental Protection Agency laboratories from July 1, 1975 to June 30, 1976, I have documented from the summaries provided by these two laboratories the following—together the two laboratories analyzed 60,665 potable water samples and 4,131 of them were positive for total coliform, in terms of percentage that's about 7½, but after verification procedures the actual percentage was about 2%. The two per cent verified positive for coliforms may have been spurious results due to a fluke in sample collection, an error by laboratory or a miscue by water treatment plant personnel.

In the *Statement of Basis and Purpose for the Proposed National Interim Primary Drinking Water Standards* (3), on page 9, there is the following statement, "The presence of any coliform bacteria, fecal or non-fecal, in treated water should not be tolerated". In the *Illinois Pollution Control Board —Rules and Regulations—Chapter 6 Public Water Supplies* (2) there is a definition of the word safe—"Safe" means that the water contains no substances or organisms which are or may be injurious to a person in normal health who ingests the water". Currently the application of check samples (4), whenever a positive coliform result is obtained whether >4 coliform colonies/100 ml by membrane filter (MF) or three or more tubes with gas by most probable number (MPN) an immediate resampling from the same sampling point is required until two consecutive daily check samples show no coliform colonies by MF or positive tubes by MPN. Thus we can assume the goal is zero coliform.

Each Environmental Protection Agency laboratory sends out procedures for collecting samples, but at times it appears to be a wasted effort as samples arrive without complete sampling data, bottles are completely full, and no one has signed the collection sheet. Before going any further, it appears we have a weak link to discuss. The weak link is the sample collector. No where in the *National Interim Primary Drinking Water Regulations* (4) is this person's responsibility described. We know in a few months the final criteria for *Water Laboratory Certification* will be published and a reference to approved laboratories is provided in Section 141.28 of NIPDWR (4). Those of us that have reviewed the proposed Criteria and Procedures for Certification of Water Laboratories Involved in Public Water Supply Analyses noticed there is a definite procedure given for collecting water samples when the laboratory is assigned the responsibility—unfortunately not all laboratory personnel collect samples. Perhaps upon rewriting of NIPDWR someone might add a statement such as this—"At the discretion of the State Regulatory Agency—water sample collectors, after a successful training period, may be issued a certificate with an identification number. Hopefully someone would compile the names and identification numbers of each sample collector for each water supply system. This would assist in a more thorough follow-up on positive samples.

Those that are quick to blame a laboratory for suprious positive results may have justification. I do know in Illinois we have more unapproved water laboratories than are approved. Also, we know even approved laboratories are capable of mistakes, cutting corners or "improving upon the method". Water treatment plant personnel or operators are capable of making an error too. But methodology today—both in water treatment and laboratory is so much better than it was when U.S. Public Health Service wrote *Drinking Water Standards*—1962.

As to maximum microbiological contaminant levels—section 141.14—suggest <1 coliform colony/100 ml by MF or <2.2 coliforms/100 ml by MPN. I see no advantage to the averaging of bacterial results if there is an exclusion of check sample results when the MCL's are exceeded. A check sample, in essence, is another sample taken from the same sampling point but does not represent the water sample that was positive for coliform and therefore necessitated the check sample. In addition to a follow-up in the water plant, there should be an examination of the sampling point to determine if the tap chosen should have been chosen to obtain a representative sample and if found unsuitable some other site in the area must be selected. A check sample is a different sample of water, and if negative for coliform would assist in lowering the average—if averages serve any purpose.

Check samples that indicate presence of coliform should be immediately reported to the water supplier and the Regulatory Agency, so the Regulatory Agency may take appropriate action. I believe the laboratories should be used to the fullest extent and their main function is to be of service. We have many commercial laboratories approved for water analysis and we feel it is their obligation to inform their clients immediately in regard to any results that indicate the presence of coliform before verification procedures or confirmation procedures are completed. The majority of our water plant laboratories do inform the chief operator of all results. However, if the present MCL's remain, the State Regulatory Agencies should be given the discretion of directing all laboratories to provide them information on sample results indicating presence of coliform whether within or exceeding the MCLs.

One of my co-workers lives in a small community of about 650 population and pays in the neighborhood of $20.00 per month for water. He feels his community water supplier should provide safe water (no coliform bacteria present), and further believes one sample per month is not adequate. I concur with him since one sample per month does not adequately monitor the water supply. This brings me to Section 141.21 Microbiological Contaminant Sampling and Analytical Requirements. I have a definite dislike the way the words monitoring and frequency are used in this section.

Sub-item b—"The supplier of water for a community water system shall take coliform density samples at regular time intervals and in number proportionate to the population served. In no event shall the frequency be less than as set forth below". Then there is the chart showing population and minimum number of samples that are required to be taken each month. Monitor as defined by Webster means to watch or check on, monitoring is the action word meaning checking on. Frequently also as defined by Webster means happening at short intervals, and frequency the fact or condition of occurring frequently. Here's the problem. We have some laboratories capable of examining all the required minimum number of samples in one day. For example, in one of our metropolitan laboratories there are fourteen analysts approved for total coliform procedures and the minimum number of samples required for them to analyze is 480 per month. If their collection unit decided to collect all samples in one day and brought them in to the laboratory, these fourteen analyists could easily analyze them. Likewise, in a smaller community of 10,600 population the Registered Sanitarian could easily collect all required twelve samples in one day and deliver them to a lone analyst in the laboratory who has the capability to process them after delivery. In both cases cited, both laboratories have the capabilities of examining all the minimum number of samples required per month in one day. Thus, as it is now written, this would be satisfactory, but it is hardly monitoring at short intervals.

I suggest the minimum number of samples per month be increased to four (one per week until all four quadrants of the community are sampled.) If only one sample is taken, the community could be in trouble the rest of month and no one would detect anything until the next month. Besides, how could those that go for bacterial result averaging do it with one sample? Not only that, it is hardly what one would call monitoring when frequency means monthly!

As of this date, our small and large community water suppliers are in a state of chaos—they all have to secure the services of an approved water laboratory or spend the monies to establish their own. A most unfortunate situation. Our state has approximately 1,650 public water systems—there are 1,348 below the 5,000 population level, 119 in the 5,000 to 10,000 level and 183 greater than 10,000 level. Of those 1,348 communities below 5,000 level, we have approximately 700 small communities in the 25-1,000 population level. Our non-community supplies number approximately 11,000-12,000. A majority of these non-community supplies are currently being sampled monthly.

Since March 14, 1977, my office has had approximately 90 requests for laboratory certification information. Many requests are from individuals wanting to start a commerical laboratory strictly for the total coliform parameters. Some requests have come from county health departments, county rehabilitation centers, factories, schools, small and large communities. Three small community representatives expressed their desire to establish a central laboratory and do samples for other communities within a 25 mile radius. They also felt sure the various mayors involved would agree to having a central laboratory with each community sharing expenses. As it appears now, there are numerous groups with the same thought, and this, in my opinion, is a feasible arrangement. As an individual community, the cost would be too great, but together it is affordable. There is also another route to go. We can establish reciprocity with our neighboring states. We have two of our commercial laboratories in Northern Illinois analyzing water samples for communities in Wisconsin and Iowa, so for the southern area of our state we will enter into a reciprocity agreement with water laboratories approved by states of Missouri, Indiana and Kentucky.

We have the technology to send men to the moon, and we have the technology to have a safe water supply.

While I decline to write a regulation, I am sure the Regulatory Engineering Staffs can. I do suggest small communities shouldn't be singled out any more than large metropolitan areas. The presence of coliform bacteria in public drinking water in either location is very undesirable. A Federal Chlorination Regulation would be of great value. We've had laboratory certification for years, but no one state has really insisted water plant laboratories obtain certification. The certification program to date as carried out by Geldreich and Nash is of the highest caliber. Those states that have approval programs only surveyed laboratories upon request—as a voluntary program. Some of us take pride in our programs and feel we have accomplished standardization of procedures so as to produce reliable data for the enforcement agencies and we've done so with the aid of Geldreich and Nash. I take umbrage that spurious positive results are solely a laboratory error and I assume the engineering staff in the water treatment plants would take offense if laboratory personnel responded with a statement "someone goofed in the plant". While the sample collector may be a weak link, I am sure they too feel resentment in being blamed. It is a matter of educating all concerned as to their respective responsibilities—to provide a safe water supply by plant personnel, proper procedures that must be used when collecting water samples, and the proper methodology that must be adhered to by laboratory personnel.

In the comments from the various regions of U.S. Environmental Protection Agency I was surprised to note a desire to give an equal ranking of MF with MPN results—I suggest a reading of *How Probable is the Most Probable Number?* by Richard L. Woodward (5), Mr. Edwin Geldreich's *Handbook for Evaluating Water Bacteriological Laboratories* (1) page 120.

For those rewriting 141.21—why is it necessary to notify the public based upon the averaging for the month? It seems to me when the first check sample is positive for the presence of coliform the State Regulatory Agency should be notified by the laboratory performing the analyses.

As for substituting residual chlorine determinations for a bacteriological sample, I suggest deletion. Bacteriological examinations detect pollution, chlorine residuals do not. State Regulatory Agencies should be given the option or allowed to use their own discretion regarding chlorine residuals, but not as substitutes for bacteriological sampling. Perhaps sample collector will determine chlorine residual at point of sampling and include information on collection sheet. Sewage treatment plants have been given the go ahead sign to use MF on chlorinated effluents and on proving results should use MPN for fecal coliform determinations. Thus coliform bacteria can be detected in the presence of chlorine.

We have no laboratories in our state using five 100 milliliter standard portions for MPN. Perhaps the use of the five 100 ml portions could be deleted.

Those of us with active laboratory approval programs have a shortage of survey forms. Mr. Geldreich's supply is very limited. Will each region allow the states to come up with the supplemental procedures survey form for SPC, Fecal Coli and Fecal Strep and permit attaching it to the federal forms that will be distributed for use? At times, it is necessary to supplement total coliform counts with results by Standard Plate Counts (SPC). SPC's are also necessary for quality control within the laboratory. Fecal coliform procedures are also used as a supplement to determine quality of source waters. The new survey forms are directed only to total coliforms. I guess I'd better go now and see the lawyer that is teaching bureaucrats to write in plain English.

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QUESTION AND ANSWER SESSION

V.J. Cabelli, U.S. Environmental Protection Agency, Narragansett, Rhode Island.

Is it advisable to have a certification program for sample collectors due to the turnover in personnel?

B.E. Gay, Division of Laboratories, Illinois Department of Public Health, Springfield, Illinois.

Yes, it is most advisable to have a certification program for sample collectors.

As each public water supply treatment plant is evaluated for requirements by State EPA Engineers and the laboratories are surveyed for compliance with Standard Methods by State Water Microbiology Survey Officers, some responsible person such as the plant engineer, chief operator or laboratory supervisor should be evaluated by the State authority for proper sample collection procedures. Upon certification by the State authority this person would then become responsible for training, surveying and certifying his own water supply sample collectors. A report of these surveys should be sent to the State EPA office, the Laboratory Survey Officer, the sample collector certification authority, and the Regional USEPA office.

This will tie in the sample collector to the program, as he may make or break the monitoring quality control program in the field.

Alternative Means of Determining Compliance

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I suppose you expect me to talk profoundly on the questions that were just proposed earlier. I don't intend to do so; I am not sure I really understand the questions, let alone the answers. I was supposed to come here and give some alternate methods to improve the bacteriological standards or replace them. Joe Harrison got here first. I therefore will say I agree with Joe's proposal No. 1, but there is one other thing I would like to see done; that is that the sampling period be a running period rather than a calendar month, for example.

If you go by a calendar month, a supply can take samples the last week of the month and the first week of the following month and then go almost two months before they have to take samples again. Or, if they should happen to miss the end of the month by one day because their bottles came late, then they are listed as not having sampled that month even though their samples came to the laboratory the following day. That is one change that I haven't heard specifically mentioned, which I would like to see made.

Probably one of the best things that I can do at this point is to try to put into perspective from my point of view, based on the responsibilities that I have, what we are faced with when we have regulations. I have heard quite a bit of talk from microbiologists; "they don't think that engineers know anything worth talking about about microbiology." If they want a perfect example it's me. So, don't expect anything really technical in that regard. But, I do have a knowledge of what happens out in the real world, which I think is quite different from what a lot of microbiologists expect. For example, yesterday I heard a lot of discussion and comment. Most of it seems to me to be directed towards a large well-run water supply with a reasonable budget and lots of money to do all sorts of things. Taking a few dozen extra samples doesn't mean anything.

We have some of those; one of the best examples is Chicago. I think Chicago is probably one of the best-run systems in the world. But we have some other types. Take, for example, a small system serving ten homes, run down, occupied by people living on Social Security. They are just barely getting along. If they have to pay the costs of samples, paying for an extra sample is a major expense. Paying for *anything* is a major expense. They don't get much, and sometimes it might be a choice between eating and getting something fixed. And, in relation to that, I think we have a very interesting Catch 22 system, "If they don't do it because they haven't got the money, we then fine them and therefore they have less money to do what they couldn't do before."

We have a fair number of these systems in Illinois. We have some that are larger than that but, up until you get to a pretty fair size you find inadequate equipment, poor operators, insufficient funds, and owners or official custodians ignorant of the technicalities of water supply. I think one of the best examples of that is when someone on my staff called the Mayor in one of the small towns to tell him that his water samples showed gross contamination, and he said, "Just a minute", came back to the phone and said, "I just drew a glass of water and it looks perfectly clear. If there is contamination there, you put it in in your laboratory." And that is a true story.

There is a lot to do in assuring the safety of a water supply, and it starts with the source. Every step is important and every step has to be checked.

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Montitoring is the last line of defense, it is really only there to see that what should have been done before has been done, and that nothing has been overlooked. So, while it is important, I wouldn't say it is the most important thing that has to be done. I don't think any of the parts are more important than another. They all have to be done and done right if we are to assure the safety of a water supply.

I suppose we are no different than other State agencies. We don't have enough money and enough people to do the job the way we think it should be done, but we do the best we can and, because we can't make all the engineering evaluations we would like to, and send our people out to all the investigations we would like to, we have to rely quite a bit on the monitoring more than I think we really should. But that is one of the facts of life and we have to live with it.

Now, getting to the monitoring itself. The ideal for a test would be something that could be run accurately by an operator, would give a definite—let's say binary—result, either yes or no, nothing in between. He would then know just what to do. He wouldn't have to use his judgment. We don't have that. Instead we have the coliform test. As we all know, this is not perfect; it has its problems. We spent most of two days here trying to discuss how to interpret the results, so that it is obvious that there must be judgment with the knowledge of the system, with the knowledge of the people who are operating that system, and of any other conditions which may affect it. It can't be done by the ordinary operator.

One of the things we can't do, as you all know, is shut off a supply. That is a last resort. We must try to keep the supply in operation and yet make it safe, which obviously is the reason for boil orders. However, this in itself creates problems because if we know that a system can be contaminated, then how can we keep it operating? If we issue a boil order, we have problems with compliance. We know people don't boil water. We had a case where we notified a supply that their water was contaminated and a boil order should be issued. The water superintendent said, yes, he would see that it was done. We checked back a couple of hours later and it hadn't been done. We told him to issue it immediately and he said, "Yeah, okay." We checked back an hour later and found that he still hadn't issued it; he was going to wait until the next day. We gave him a few choice threats, and insisted that it had to be done immediately. He went on television and said, "The Illinois EPA says you should boil your water, but I don't think you have to and I am not going to boil mine." That is almost an exact quote.

When we take a sample we hope that it is representative. But really it is only representative of the water that is passing by the sampling point at that instant. We will never be able to get a representative sample of that particular water again because as it goes through the distribution system, it changes and it mixes with other water in the system, is dispersed, so the sample we get is a sample of one discrete portion of water. However much we might try, it is not representative of the whole system and never will be. We have to take that in consideration when we set a standard.

We have to be very careful when we have public notification. I can give you an example in a slightly different area. We have mandatory fluoridation in Illinois. I had a woman call me up to ask me if there was some way in which a petition would get her municipality released from mandatory fluoridation because she didn't want it. I talked with her for a little while and found out that she was drinking bottled water. They were buying bottled water for drinking purposes—she, her husband and the children—and they were only using the public water supply for bathing and so forth. We got a sample of the brand of bottled water that she was using. The municipality was holding pretty well between .9 and 1.2 parts of fluoride. The bottled water had about 5 parts.

When people start distrusting their water, they'll go elsewhere, and almost invariably the other source will be less safe than the public water supply. If we start giving public notice with little or no real backing for the notice, in other words, if we give it just because a number has been exceeded (a number which has been selected somewhat arbitrarily), then we are going to have two results. We will probably have first one, and then the second. The first result will be panic and will have people going off in all directions trying to avoid the use of the water at all costs, including using the nearby pond, and that stream that looks so nice and clear as it runs past the sewage plant outfall. Then, they are going to decide that this is just a lot of crying of "wolf", and after a while, when we have a major emergency, we will send out a notice and nobody will pay attention. And that is one of the things that I am worried about. I would hate to see that attempts to implement the Safe Drinking Water Act would eventually result in poorer or less safe water for the communities in this country.

Thank you.

Alternative Means of Determining Compliance

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Over a period of a day and a half it has become quite clear that there is a real problem associated with establishing microbiology standards for drinking water. What is not clear is the exact nature of the problem. It may be a public health problem. It may be a microbiology problem. It may be a statistics problem. It may be the engineering problem of providing adequate treatment facilities and distribution systems whose integrity is difficult to broach. It may be the personnel problem of finding capable people and training them properly as operators, water samplers, and laboratory technicians. It may be a regulatory problem. It is probably a mixture of all of these.

Previous speakers have concentrated mostly on the technical parts of the overall problem. Thus, it is appropriate for me to make a few comments on the nature of the regulatory problem.

First, I would like to say that I have never seen a technical problem solved by a regulation. Technical problems cannot be legislated (or regulated) out of existence. They have to have technical solutions if they are to be solved at all. However, regulations can create technical problems. There needs to be some very serious thought about separating the regulatory part of the overall problem from the technical parts and about how to word the regulation so that it motivates efforts to solve the technical parts of the problem.

There is a basic conflict between regulatory approaches and technical approaches. Lawyers write regulations and we can be sure that whatever subjects and numbers technical people put into the regulation to begin with, the lawyers will have the last chance to determine the wording. Lawyers talk about "good draftmanship". What they mean is that in wording a law or a regulation it should be made clear that, if a person does A, B, and C, he is operating perfectly legally and within the law, but if he does A, B, C, and D he has violated the law and is subject to a fine or a prison term or whatever other penalty they can dream up. They want to make the penalty of going from C to D so high that few people will take the risk.

In technical matters, usually A is excellent, B is good, C is poor, D is bad, E is worse, and F is awful. Somewhere within the range of C, D and E, a line between the acceptable and the unacceptable is passed but the exact location of this line is difficult to determine precisely. As an example, if the monthly average concept is used in the microbiology standards for drinking water, a specific number needs to be selected. That number may be 1 coliform per 100 ml or 2.2 per 100, or 4 per 100. All of these numbers are in the gray area between acceptable and unacceptable and probably none of them is technically defensible in an absolute, quantitative sense. If any one of those numbers is selected, it is quite likely that someone will come up with a number of examples of water supplies which exceed the monthly average standard in communities where there is no evidence of waterborne infectious disease. On the other hand, an average of 16 coliforms per 100 ml is probably far over the black and would be running too great a risk of outbreaks of waterborne disease in too many communities.

The Primary Drinking Water Regulations are written in terms of maximum contaminant levels (MCL's). It appears that there is some attempt to apply the MCL concept to different types of parameters on a somewhat uni-

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form basis. This may be a source of part of the problem. Water treatment processes are not in particular designed to remove arsenic, barium, cadmium, chromium, lead, mercury, nitrate, selenium, or silver although some amount of some of these substances may get removed during treatment. Water treatment processes are in particular designed to remove turbidity and coliform bacteria from water. There is a statement in *Standard Methods* (1) which says

> ". . . no coliform bacteria of any kind should be tolerated in a treated water."

This statement is intended to differentiate between the use of the coliform group and fecal coliforms as indicators rather than to establish a drinking water standard and it is an extreme viewpoint. However it does emphasize very well the concept that treated water should have no coliforms in it.

A properly operated treatment system usually turns out finished water with no coliforms present. When there are no coliforms in the finished water, any coliforms found in the distribution system indicate that water from some other source has been introduced. The discovery of coliforms in either the finished water or in the distribution system indicates that something is wrong and corrective action should be taken. If coliforms are found in the finished water at the treatment plant, the treatment system should be investigated to find out why they are getting through and adjustments should be made so that they don't. If coliform organisms are found in the distribution system, the source of the water containing coliforms should be located and eliminated. On the other hand, if the positive coliform tests are the result of poor sampling or poor laboratory technique, this needs to be discovered and corrected. Regulations written in terms of monthly averages and percent of samples showing positive results just don't get the message across; they infer that it is O.K. to have some coliforms present. The regulations should be written so that it is clear that positive results are not O.K.; they indicate a problem that needs to be solved and needs to be solved promptly.

Testing finished water at a well run water treatment plant for coliform bacteria is not a very rewarding activity because practically all the results are 0 coliforms per 100 ml. That is why my research to determine the frequency distributions for coliform organisms in water has been done using raw water samples. The frequent result of 0 coliforms per 100 ml is a reason to reconsider the use of monthly averages in the regulations. That result doesn't mean that there are no coliforms present; it just means that the sample was too small. If a liter, or 10 liters, or 100 liters, etc., of water were filtered, sooner or later a coliform organism would turn up. The 0 coliforms per 100 ml just indicates less than 1 coliform per 100 ml. Averaging a string of numbers which include several zeros which really aren't zeros creates problems which the statisticians haven't solved yet.

There are statistical problems in estimating the number (or range of numbers) which best represent the results of a series of coliform determinations. I say this in full knowledge of the work of Clarence Velz during the 1940's (7) and Harold Thomas during the 1950's (6). Their work really was excellent considering that they were sanitary engineers doing statistics. The questions we are asking now are somewhat different, we are demanding more precise answers, and we have more serious and more economically far-reaching decisions to base on those answers. Also the field of statistics has seen several new developments in the last 15 years and some of the newer statistical techniques offer better answers to our questions if we had the right kind of data.

Dr. Litsky presented a strong indictment of the proposed coliform standards based on the lack of precision of the most probable number (MPN). What he presented is correct; however, he didn't give a fair and balanced presentation. It is true that McCrady (5) was the first to use the term, "most probable number", and it is true that he used Bayes' concept of inverse probability in his derivation of the MPN equation. It is also true that the Bayesian approach has been discounted by mathematicians and no statistician would use that approach now. McCrady was a bacteriologist not a statistician and his entire mathematical approach was crude. However, the same MPN equation was derived more elegantly by Greenwood and Yule (4) two years later. It was also derived by R. A. Fisher (3) in his classic paper on parameter estimation in 1921 and there are several other derivations of the MPN equation which have been published since. I think that we have to accept the mathematical soundness of the MPN equation if we accept the assumption on which it is based.

The term, "most probable number," is an unfortunate one because statisticians have rejected the Bayesian concept of inverse probability and because the term implies a precision the number doesn't have. It should be called the "maximum likelihood estimator of the mean coliform density" but, of course, McCrady didn't know this when he wrote his paper because Fisher didn't invent the term "maximum likelihood estimator" until six years later. It is remarkable that a bacteriologist anticipated Fisher's general result for this specific case by six years.

The other references which Dr. Litsky cited were trying to point out that the precision of the MPN is not as high as most people who use the multiple tube fermentation test think it is. They were not saying that the MPN value had no precision at all. It would, of course, be a good thing for the people who use MPN values to understand what its precision actually is and for this to be taken into consideration in the regulations.

Standard Methods (1) does give the 95% confidence interval range for MPN values. This range in most cases is a factor of 5; that is, using the table in *Standard Methods* we can distinguish between water with 1 coliform per 100 ml and water with 10 coliforms per 100 ml but we can't distinguish between water with 1 coliform per 100 ml and water with 5 coliforms per 100 ml. The table in *Standard Methods* is a bit optimistic. The assumption behind the MPN equation is that the numbers of bacteria in small volume aliquots from a water sample will fit a Poisson distribution. The characteristic of the Poisson distribution is that the variance is equal to the mean. Anyone who has had much experience doing coliform counts on water samples knows that the variance is much greater than the mean. It even says so in *Standard Methods* (1). That was the main point Dr. Muenz was trying to make yesterday when he was discussing the data I had sent to him.

The statistical model which Dr. Muenz developed is really rather simple compared with models other statisticians have suggested to me and it is very powerful. If we had the right data on coliform counts in treated water and water in distribution systems his model could be used to put the sample frequency table of the regulations on a rational basis. I suspect that his "bowling ball" model; that is, most of the water with no coliforms and only an occasional slug of water with some coliforms is the correct one. From that, I suspect that if the sampling frequency table were put on a rational basis, it would indicate a need for a much greater sampling frequency than anyone has suspected so far.

I have talked mostly about the regulatory problem and the statistical problem because the other technical aspects of the overall problem have been well covered by other speakers. I would like to end by agreeing with a couple of previous speakers.

I agree with Lee McCabe about the need for real time response to bacterial contamination of a public water supply. Such a contamination needs to be detected as soon as possible and needs to be corrected as soon as possible. The bacteriological tests we have are not suited for a monitoring program because of the turn around time. Some parameter such as residual chlorine needs to be used for monitoring purposes. It would be more rational to use the microbiological testing for short-term, intensive studies of the level of contamination of the raw water, of the effectiveness of the treatment system, of the integrity of the distribution system, and of the validity of some alternate monitoring system.

The last thing I want to say is that I agree with what Archie Greenberg said this morning about flexibility in the regulations. Coliforms are surrogates for pathogens or for public health effects. Standards and regulations are surrogates for judgments. The regulations attempt to prejudge every situation which might arise. We just aren't that smart. A regulation might cover 90% of the cases adequately but there always will be exceptional cases in which following a regulation literally will lead to the wrong response to the situation. The regulation must be written with enough flexibility so that the technical people who have the direct responsibility for water supply can find the right response and solve the problem.

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Alternate Means of Measuring Compliance with Bacteriologic Standards of Public Water Supplies

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The question of what constitutes an adequate measurement of compliance with bacteriological standards in water supplies should be extended to a consideration of the standard itself and of its utility for the protection of public health. For the purpose of this conference, therefore, we should attempt to develop a logical connection between the absolute measurement of performance by bacteriological testing and the meaning of that performance in terms of public health.

In an earlier paper (1) I attempted to define mathematically the relationship between bacteriological standards and public health risk for bathing beaches. A distinction was made between two situations, governed by what might be called the laws of large and small numbers. These lines of reasoning are easily applied to considerations of water system safety.

The terms *large numbers* and *small numbers* refer to the size of the human population in a watershed. For sources of potable water polluted by large populations (e.g., the Great Lakes and the Mississippi and lower Hudson rivers), the probability of ingesting an infectious dose can be calculated from the probability of ingesting a certain number of viable infectious particles and the probability of infection from such a particle. This calculation must include proportonality factors, such as (*a*) the abundance of excreters of infectious particles in the human population and (*b*) the ratio of infectious particles excreted to the number of indicator bacteria. A large polluting population virtually assures the presence of the infectious agent, but in numbers that are reasonably stable and predictable and generally quite low.

When water polluted by a large population serves as a source of potable water, the public health risk associated with its use depends to a great extent upon the abundance of excreters in the watershed population, which determines the ratio of infectious particles to indicator bacteria. The density of the infectious agents can be reduced, often by several or many orders of magnitude, by precipitation, filtration and an initial disinfection step. For infectious agents which can be effectively removed by this treatment, the product will often be quite safe for drinking even without final disinfection—not because the pathogens are absent, but because their numbers are so small as to virtually preclude ingestion of an infectious dose by any one consumer. Final disinfection can then be considered mainly protection against sources of contamination affecting the treated water.

The law of small numbers applies to sources of drinking water affected by small populations and to drinking water distribution systems. Whenever the polluting population is small, the ratio of pathogens to indicator bacteria is subject to wide fluctuations depending on the presence or absence of excreters in the watershed. The safety of the supply with regard to a specified disease is assured when there are no excreters in the population, but the water may be highly unsafe if and when only a few or even one excreter is present. In the extreme case the probability of one excreter being present at any time is virtually identical with the probablity of an outbreak. Assum-

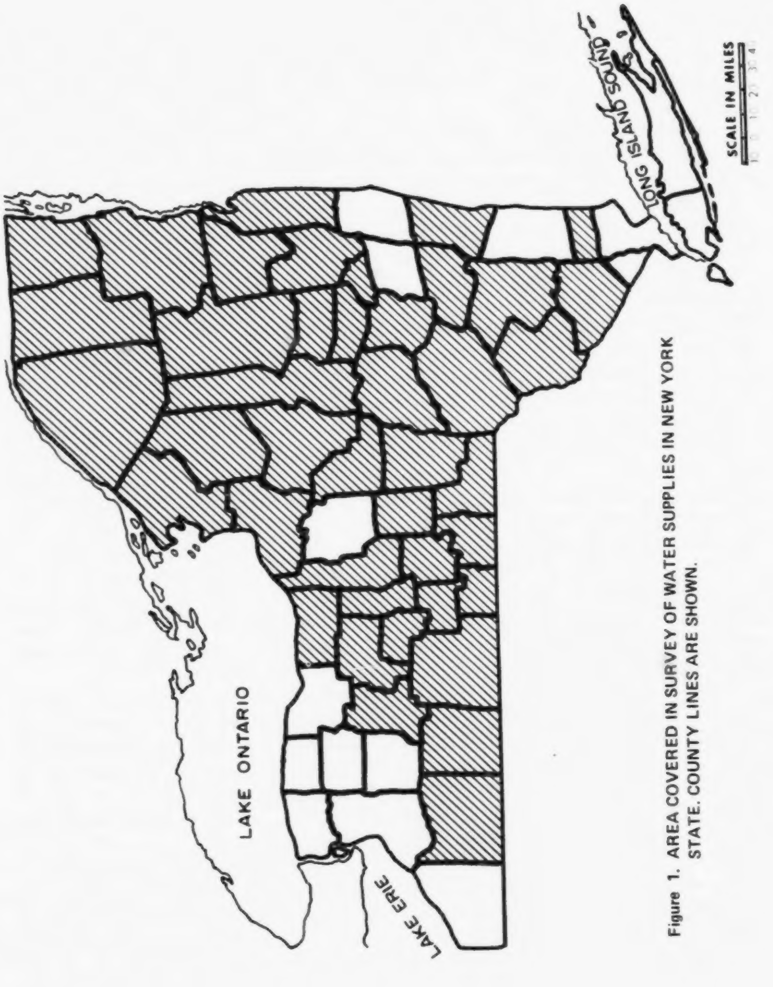
LAKE ONTARIO
LAKE ERIE
LONG ISLAND SOUND
SCALE IN MILES

Figure 1. AREA COVERED IN SURVEY OF WATER SUPPLIES IN NEW YORK STATE. COUNTY LINES ARE SHOWN.

ing that excreters are randomly distributed and as mobile as the general population, this probability will obey a Poisson distribution with time, with the ratio of excreters to general population as the mean. Therefore the public health risk in this case also is predicated upon the prevalence of infection in the general population.

This principle explains three phenomena: (*a*) short episodic outbreaks of waterborne disease related to occasional pathogenic contamination of a water supply whose treatment system fails intermittently or contiuously, (*b*) the absence of waterborne disease in certain small, isolated communities, despite almost constant contamination of their water systems with fecal bacteria, and (*c*) the occasional water system which produces illness whenever a vital disinfection step fails, indicating access to the system by a resident excreter.

In dealing with situations governed by this law of small numbers and for the purpose of *protecting* (rather than *predicting*) public safety, the number of excreters for any given disease in the population should be abitrarily set at the theoretically expected number *plus one*. This additional excreter represents the individual who may have access to the system at some time in the immediate future and thereby create an acute hazard. This approach, in effect, leads to a policy of zero permissible contamination for all potable and recreational waters affected by small populations and, in particular, for all public water supplies, since their distribution systems are susceptible to localized access or failure.

The strategy for a zero-contamination policy consists in setting up one or several protective barriers: watershed protection with fences and patrols; impermeability of sources, reservoir structures, and distribution systems; and disinfection. Each barrier is intended to be absolute but is in fact not so, due to the imperfection of all human endeavor. The effect of any small number of failures of any barrier can be reduced, often dramatically, by a second protective barrier, as long as the failures of either barrier are randomly distributed with time and their causes are independent of one another. In this case, the over-all probability of failure of the barrier system is the product of the probabilities of failure of the individual barriers (together with the probability that some person having access to the system is excreting pathogens, if fecal bacteria are the indicators of system failure).

Since these probabilities combine in a multiplicative fashion, we should not be surprised to find a lognormal distribution of the number of incidents of contamination with fecal bacteria per unit of time. This indeed is the case.

Bacteriologic records from 726 public water supplies in 42 mostly rural counties of New York State (Fig. 1) were examined for reports of coliform bacteria. (This is the geographical area served by the sanitary bacteriology laboratories of the New York State Department of Health.) Detection of any coliform bacteria in one or more samples in any calendar month during which samples were received was counted as one occurrence. This was done in an attempt to assign equal weight to failures of the source, of the treatment, and of the distribution system, although the use of the calendar month is rather arbitrary. Most records covered 3 years (July 1972-August 1975), with a minimum of 12 months. Sampling frequency was based on population according to the 1962 Public Health Service table, but with a minimum frequency of 12 rather than 24 per year.

The cumulative frequency distribution of the average number of occurrences per year, plotted on log-probability paper, shows a straight line over most of the range (Fig. 2). This reflects the multiplicative behavior of component probabilities, as explained above. The different slope of the curve in the range of supplies which fail most frequently may indicate a different underlying mechanism: these are probably supplies which are continuously contaminated, often at a low level, so that the contamination fails to appear sometimes in the single 100-ml samples tested.

Figure 2 shows that 17% of the supplies were free of coliforms for 3 years; the median value was 1 event every 13 months, or 0.9 events per year. However, 10% of the systems showed coliforms every 3 months or more frequently, and 1% were more or less continuously contaminated. Typical examples of poorly performing systems are (*a*) a supply with a defective branch in the distribution system which continuously delivers contaminated water to an outlying section of the community and (*b*) defective small supplies in economically very depressed regions where improvements cannot reasonably be expected without financial assistance from the outside.

This method of determining the performance of a water supply is much more stringent than the existing regulatory procedure, which allows the

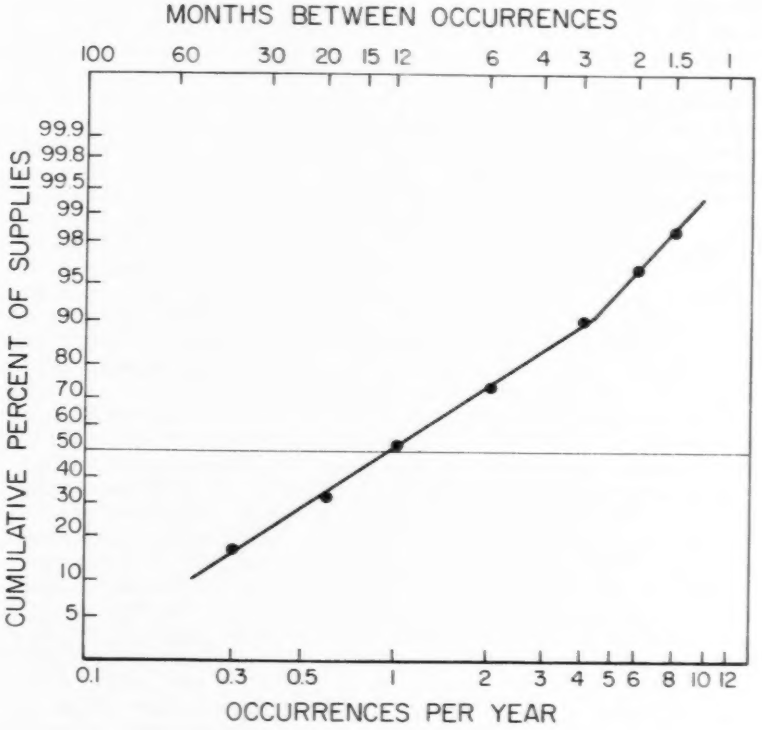

Figure 2. CUMULATIVE AVERAGE FREQUENCY DISTRIBUTION OF COLIFORM OCCURRENCES IN NEW YORK STATE WATER SUPPLIES. FOR DEFINITION OF OCCURRENCE AND PERIOD COVERED, SEE TEXT. FOR AREA COVERED, SEE Figure 1.

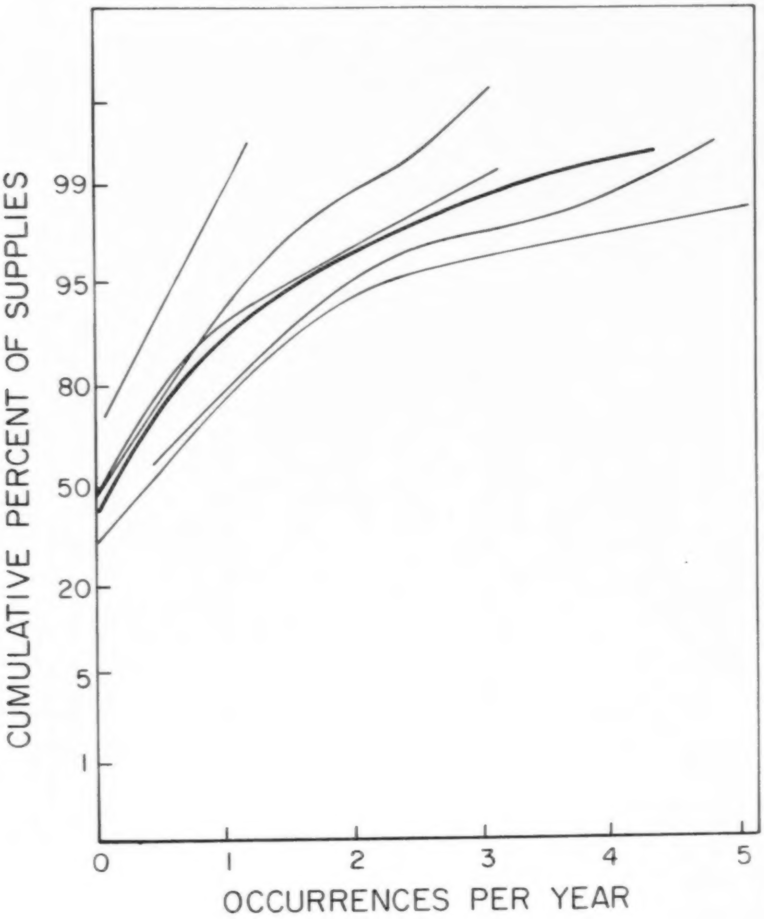

Figure 3. CUMULATIVE FREQUENCY DISTRIBUTION OF COLIFORM OCCURRENCES IN FIVE LOCAL JURISDICTIONS IN NEW YORK STATE. THE SUMMARY CURVE (HEAVY LINE) APPROACHES THE CURVE OF THE LOGNORMAL DISTRIBUTION IN Figure 2.

averaging of coliform counts over all samples taken in any given month. It also is suitable for characterizing the status of an entire region or jurisdiction. Differences among five local jurisdictions in northeastern New York State are illustrated in Figure 3. While the summary curve approaches the lognormal distribution, the individual curves show markedly different degrees of performance, which are suggestive of both regional socioeconomic differences and the varying intensities with which corrective actions are sought on the local level.

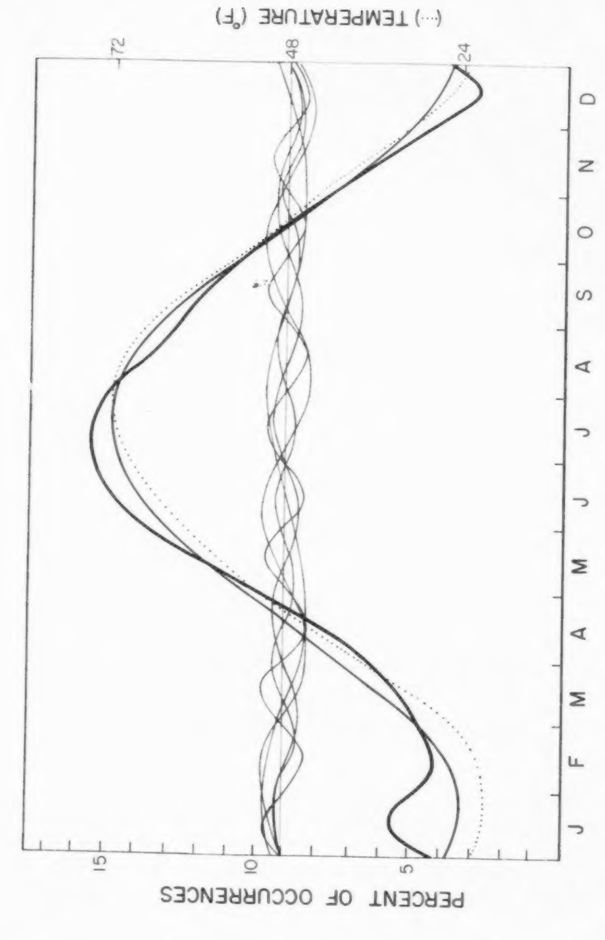

Figure 4. SEASONAL DISTRIBUTION OF COLIFORM OCCURRENCES IN THE FIVE JURISDICTIONS. HEAVY LINE: ACTUAL OCCURRENCES. LIGHT LINES: CONSTANT, FUNDAMENTAL, AND FIRST FIVE HARMONICS OBTAINED BY FOURIER ANALYSIS. DOTTED LINE: LONG - TERM MEAN MONTHLY TEMPERATURES FOR ALBANY, NEW YORK. THE DISCREPANCY BETWEEN ACTUAL EVENTS AND PREDICTIONS IN DECEMBER AND JANUARY WAS AN ARTIFACT OF THE SAMPLING SCHEDULE.

The 726 supplies in our sample produced an average of 650 occurrences per year, or almost 2,000 occurrences over the 3-year period covered by this study. Yet there were only two outbreaks of disease. One occurred in a larger supply which had about 4 occurrences a year but was mostly in compliance by regulatory standards because these are based on average counts, which in this supply were generally low. The outbreak was ascribed to waterborne *Giardia lamblia*. The other outbreaw of disease was of a particular freaky nature and can hardly be called waterborne in a strict sense. It was ascribed to a rare pathogen, *Yersinia enterocolitica*, which was traced to a supply with an almost flawless bacteriologic record. The supply itself did not carry an infectious dose for any consumer, but the bacteria multiplied in a commercial dairy product, causing the outbreak. This low ratio of outbreaks to system failures shows that the pattern of outbreaks of waterborne disease is governed by the laws of probability.

The seasonal distribution of coliform occurrences in the five local jurisdictions covered in the previous sample is shown in Figure 4. Fourier analysis (the first six harmonics are shown) indicates a seasonal pattern which closely follows the long-term mean monthly temperature curve for the area. The abnormality in December-January is an artifact caused by the sample collection schedule.

Another important consideration in formulating regulations on bacteriologic sampling and in determining compliance is the statistical power of the bacteriologic test. The most powerful statement that can be derived from the standard tests is the proof that coliforms (or any other selected group of bacteria or agents) were present in the sample and therefore were present in the system at the time of sampling. Whenever we deal with small numbers of bacteria in a small number of samples, the quantitation of a positive result is characterized by poor precision. But the power of the negative test is also quite poor. A negative result for a single 100-ml sample, if interpreted at the 95% confidence level (which is standard in scientific work), should read "fewer than three bacteria per 100 ml" and not "fewer than one," as has been customary in water supply monitoring. The probability that the statement "fewer than one" is correct is only about 70%. (The remaining percentage points are attributable to the possibilities that there are one or two bacteria per 100 ml.) Even this statement, which is based on the Poisson distribution, holds only for small parcels of water, which are likely to show a random and homogeneous distribution of bacteria, and does not hold for entire distribution systems.

Whenever the statistical power of a single test is small, we are told by statisticians to continue testing until a satisfactory conclusion can be reached. This is exactly the purpose of the long-term evaluation shown in Figure 2. As long as a water system does not undergo a significant change the record reflects "present conditions" except for seasonal variations and random events. If the sampling period spans at least 1 year and if at least 24 samples were taken, a rating of the system by this method is possible. I tend to prefer a series of monthly samples taken during 3 years over the same number of samples taken more frequently during 1 year because the 3-year series may include events such as floods, droughts, or other unusual stresses on the water supply.

This type of long-term evaluation has the advantage that a single occurrence of bacterial contamination is placed into proper perspective. Individual occurrences of bacterial contamination may be ascribed to unusual circumstanes and may be excusable, but a high rate of recurrence of such incidents is an indication of an unreliable source, ailing equipment, sloppy operation, or any combination of these and other factors. The single incident also rarely produces an outbreak of disease, but the aggregate of all incidents is a direct measurement of the tightness of the system and therefore of the public health risk associated with it.

In this context I feel strongly that *no positive bacteriologic results should be discarded without cause*. In our experience, isolated positive results tend to become confirmed in the long run, although not necessarily in the check samples immediately following, which may not represent the same parcel of water. Also, in our experience, false-positive results by laboratories are less frequent than false-negative results, but this may in part be due, not to laboratory error, but to the low statistical power of the negative result, as explained above.

Long-term evaluation of the type indicated can be used to set priorities for a detailed investigation of systems that are performing poorly. Such an investigation would include sanitary surveys with intensive sampling, using a more complete spectrum of bacteriologic and chemical techniques. These should include nonstandard but nevertheless firmly established microbiologic procedures for the identification of bacterial types indicative of specific sources of contamination. (The summary methods of coliform determination and standard plate counts are appropriate for determining the presence or absence of any number of problems, but they do relatively little to reveal the exact nature of the problem.)

The detailed investigation of poor or marginal supplies should be directed at finding the most effective ways of upgrading them. The investigation of a supply with average performance should aim at determining whether a significant problem exists and whether performance could be improved at a reasonable cost.

Long-term evaluation and investigation are directed at finding ways for long-term improvement of water supply operations and long-term improvement of the prevalence of infections in the general population, which, as we have seen, is essential in assuring that the fewest possible system failures result in disease. Nothing should discourage regulatory peronnel from following up positive bacteriologic results immediately and taking all action necessary to protect the public from acute health hazards. But an equally important long-term goal is a conclusive rating of each water supply. This cannot be obtained by a highly variable, month-to-month rating, which only breeds confusion, indecisiveness, and, if combined with a public notification scheme, public apathy.

As the problems of many water supply systems are chronic or of a long-term nature, the solutions are often not of the quick-and-easy type. Disinfection creates only one barrier where several are needed. Chlorination cannot compensate completely for the shortcomings of deteriorating distribution systems, and the formation of chlorinated organic materials has become a cause for concern. There is need for improved watershed protection, for treatment by advanced techniques, and particularly for the rehabilitation of old distribution systems. The need for costly, long-term solutions involving construction projects is particularly obvious in areas in the northeast which are already economically depressed.

Public notification of a poor long-term and overall performance of a supply —as opposed to proclamation of an acute public health hazard—can be an important instrument to obtain support from an informed public for long-term improvement of a water supply. The prohibited-use classification of the U.S. Interstate Carrier Sanitation Program was a valuable precedent in this regard. In the absence of a construction grant program for water supplies, correct handling of the public notification issue assumes particular importance if activities under the Safe Drinking Water Act are to become more than a rather fruitless, frustrating exercise.

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QUESTION AND ANSWER SESSION

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What is the appropriate action for a one-barrier system to provide absolute safety from an outbreak of disease?

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Make sure that the barrier works at all times. If the barrier is a chlorinator, it must be watched 24 hours a day, 7 days a week.

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EVALUATION OF THE MICROBIOLOGY STANDARDS FOR DRINKING WATER

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